INVISIBLE RADIATIONS OF ORGANISMS

OTTO RAHN

HERITAGE

INVISIBLE RADIATIONS OF ORGANISMS

BY

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WITH AN INTRODUCTION TO THE PHYSICS OF RADIATION

BY

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WITH 52 ILLUSTRATIONS

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FOREWORD

Visible biological radiations have always greatly attracted man's curiosity; fireflies and glow worms, luminescent wood, phosphorescent meat, and the illuminating organs of deep sea fish are among the well-known wonders of nature. The biological importance of this luminescence seems to be in no proportion to the impression it makes upon the human mind. While it is assumed by some biologists that it has the purpose of attracting the prey, of frightening enemies, or of luring the male to the female, other investigators have contested these theories. The phosphorescent bacteria usually lose the ability to produce light when cultivated for some time on artificial media, without any apparent decrease in vitality. The emission of visible light is probably of no greater importance than color; it plays no essential role in the cell physiology of the organisms.

Quite to the contrary, the invisible radiations of living organisms are of considerable physiological significance. They play a distinct part in cell division and in growth. They are evident in the healing of wounds. Old age is accompanied by complete cessation of ultraviolet emission; perhaps this is the cause of old age. *Beta*-radiation controls the heart beat. The loss of blood radiation is used in the diagnosis of cancer; it may be that radiation, or some disturbance of its mechanism, is linked with the cause of cancer. Its role in the metamorphosis of amphibia has been demonstrated. Mutual influences of one species upon another by radiation have been observed.

Biologists and physicists have always been suspicious of radiations from living organisms, perhaps only because the average man (not to mention woman) likes to believe in human radiations. However, the principal reason for the rejection of the discovery of ultraviolet emission from living cells was the inability of some to repeat the positive experiments of others with the same results. This had led to the fallacy that negative results disprove positive ones. It is quite evident that if two experimentors obtain different results, they cannot possibly have made the same experiment. Both results are correct, and the important task is to find out in what points the investigations differed. With a phenomenon so little understood as these biological radiations, it is not surprising that these apparent contradictions have not as yet been explained in every case, though several factors responsible for negative results have been discovered.

The objection to biological radiations has been strongest in this country, but even here, a more conciliatory attitude has become noticeable since it has been shown that mitogenetic radiation is not a mysterious force, but the result of biochemical processes. Many simple chemical reactions have been found to emit weak ultraviolet rays. Another factor is responsible for the slow adoption of this new influence in biology: practically all papers on this subject are published in foreign languages, and of the very few in English, almost all happen to contain negative results. This very fact has been one of the authors' reasons for presenting the more important facts in this book.

The book deals almost exclusively with mitogenetic rays which exist in the ultraviolet range of the spectrum. No definite proof for the emission of infrared rays by organisms could be found (if we limit the infrared to radiations near that of the visible). *Beta*-ray emission from potassium is biologically important, but it is not really characteristic of the living cell; it is proportional to the potassium content, and is just as strong after death as during life.

The arrangement of the subject matter is not historical, but logical. An attempt has been made to show that ultraviolet radiation from living organisms is nothing at all strange. If GURWITSCH had not discovered these emanations 10 years ago, they would now be predicted from the results of physico-chemical investigations. An approach to historical presentation is found in Chapter IV which discusses the various methods used.

The book is not meant to represent a compilation of all literature on this subject. This would have increased its size greatly. A fairly complete list of references, up to the beginning of 1932, may be found in the book by STEMPELL (1932). The literature compiled at the end of this book refers only to those papers which have been quoted in the text; we suppose that this includes the more important publications.

A very brief summary of the entire book, chapter by chapter, is given at the end. This might be more useful in some respects than the customary Table of Contents.

One of the authors had occasion, during a recent journey to Europe, to see many of the biologists and physicists working in this field, and he wishes to acknowledge the many valuable suggestions he received from those convinced of mitogenetic radiation as well as from those who are convinced of its non-existance. The authors are further under great obligation to Professor ALEXANDER GURWITSCH for sound advice on various points, and to Professor MAGROU for the kindness of sending original photographs of his experiments for the reproduction in this book.

The authors are further under great obligations to Mrs. MARGARET N. BARNES for her ceaseless assistance in editing this book, and to Miss A. J. FERGUSON for her help in proof-reading.

Ithaca, August 1935

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SIDNEY W. BARNES

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CHAPTER 1

PHYSICS OF RADIATION

A. GENERAL STATEMENTS

(1) Radiation travels through empty space. From the studies of radiant energy have come several ideas about its nature. For one thing, it is known that such energy is propagated through empty space. Moreover, it is the only form of energy known which can flow through matter-free space. The vast amounts of solar radiation which maintain the earth at such a temperature that life is possible, come from the sun through millions of miles of interstellar space, which contains but an infinitesmal density of matter.

(2) Radiation energy spectrum. Radiation occurs over an extended energy range. The extremely high-energy *gamma* rays penetrate several inches of lead; the lower-energy *x-rays* pass through perhaps an eighth of an inch of lead; *visible light* is absorbed by a metal layer only a few atoms thick; and, finally, *radio waves* are completely absorbed by a coarsely-woven copper wire screen. The character of radiation varies greatly, as one can see, from one part of the radiant energy spectrum to another.

(3) Radiant energy travels through space with a fixed, definite velocity. Radiant energy might be given the alias of traveling energy, for it spends each instant of its existence traveling through space at its particular speed of 3×10^{10} cm. per second (speaking here of space in which the matter density is zero). This speed is entirely independent of the character of the radiation. To the best of our knowledge, radio waves, visible light and gamma rays all travel with precisely this velocity. The direction of travel is rectilinear.

(4) Radiation exhibits the phenomena of interference. Any form of wave motion can be made to exhibit the

phenomena of interference. The beats heard when two tuning forks of nearly the same frequency are struck, are an example. Later (see p. 128), an example of interference exhibited by light will be given.

(5) Radiation may be observed when, and only when, it is allowed to interact with matter. This statement is a reminder that all recognized measurements of energy are limited to energy associated with matter. To detect or measure radiant energy, it is necessary that it be transferred into the familiar potential or kinetic energy of matter. This transformation obeys the law of conservation of energy, i. e. the number of ergs of radiant energy disappearing is equal to the number of ergs of potential or kinetic energy which appear.

B. PHENOMENA OBSERVED UPON THE INTERACTION OF RADIATION AND MATTER

(1) Reflection. Radiant energy may be regularly reflected from a plane surface whose granular structure is small in comparison with the wave length of the radiation. In this case the angle of reflection is equal to the angle of incidence, and the reflected energy is in the plane of the incident energy. No perfect reflectors of radiation are known; thus, always some of the radiation is transmitted or absorbed by the reflector.

(2) Absorption. Matter never fails to take its toll from the radiation incident upon it. No material substance known is totally transparent to radiant energy. The mechanism of absorption will be treated later (see p. 16).

(3) Refraction. Matter has the property of changing the velocity of energy which is passing through it. This results in a change of direction of the radiation (see fig. 1). The ratio of the velocity of radiation in matter to the velocity in space is called the index of refraction of the refracting substance.

(4) Dispersion. The phenomenon of dispersion occurs because the index of refraction of any transparent material depends also upon the wave length of the radiant energy which is passing through it. A beam of white light is dispersed into a colored band or spectrum when passed through a prism since each wave length suffers a different refraction (see figs. 2a and b; also see p. 18).

These are only a few of the many phenomena which characterize radiant energy in its passage through space and matter; these must be explained by any theory of radiation. The question: *What is the nature of radiant energy*? has been nearly answered by each of two different theories, the wave theory and the quantum theory. As it will be pointed out later, it seems not impossible to effect a harmonious combination of these two into one which adequately covers all the observed phenomena of radiant energy.

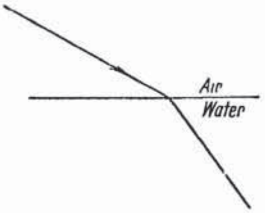

Figure 1. Refraction.

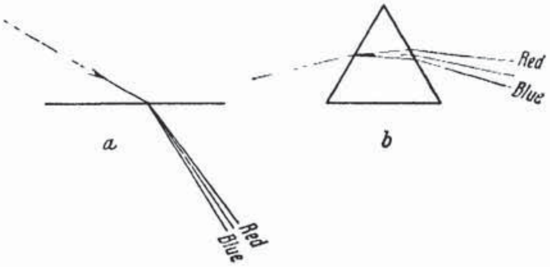

Figure 2. Refraction and Dispersion.

The so-called "mitogenetic radiation" which is the principal subject of this book is said to proceed rectilinearly, and to show reflection, absorption, refraction and dispersion, as will be demonstrated in Chapter IV. If so, it is a true radiation.

C. THE WAVE THEORY OF RADIANT ENERGY

There are three ways in which energy may be transferred with the aid of matter, namely (1) by the flow or movement of definite masses of matter, such as tides in the seas, or the drive rod on a locomotive; (2) by wave motions in elastic media, such as sound in air; and (3) by material projectiles. The wave theory of radiation is based upon the well understood principles of wave motions in elastic solids. These may be illustrated by the following simple experiments.

If a long stretched rope is given a blow at one of its supports, a rather surprising thing happens (at least so to the uninitiated); a hump in the rope is seen to speed along it. If, for a short period of time this end of the rope is given a regular to-and-fro motion, a disturbance as pictured in fig. 3 will travel along it with the same velocity as in the former case. This sort of disturbance is called a wave train. The length of the individual waves, or the wave length, λ, is the distance from crest to crest or trough to trough. The frequency, ν, or the number of waves passing a fixed point per second, is equal to the velocity, c, divided by the wave length:

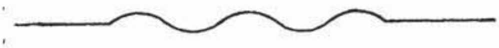

Figure 3.
A short wave train in a rope.

$$\nu = \frac{c}{\lambda}.$$

Reflection. Let this wave train be observed when it reaches the end of the rope. If the support there is ideally rigid, the train of waves will be reflected and will travel back along the rope with the same velocity and amplitude it had before reflection.

Absorption. If the support is ideally non-rigid, the wave train will set it in motion, spend its energy upon it and completely disappear. In this event, the energy carried by the wave train has been transferred to the support.

Standing Waves. There is still another phenomenon of wave motion which may be illustrated by waves in the rope. When it is fastened to the rigid support, if the free end is kept moving with a uniform to-and-fro motion, so-called standing waves will be formed (see fig. 4). The rope appears to be divided into vibrating segments called *loops* which are separated by points of little or no motion called *nodes*. These standing waves, not true waves at all, are the result of the interference of the original

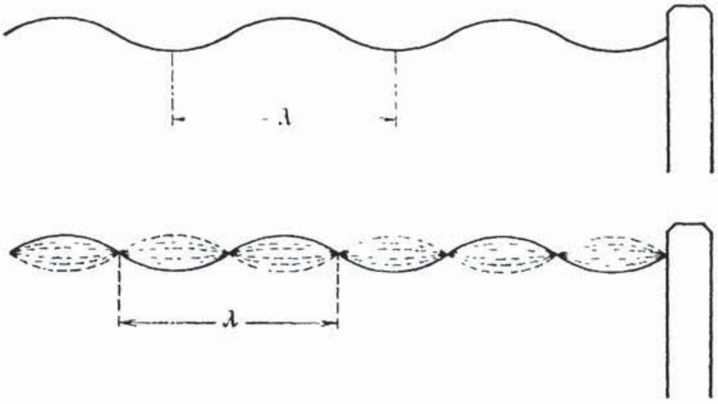

Figure 4. Above: Waves in a rope; below: standing waves in a rope.

and the reflected waves. Their importance lies in the fact that they offer a very simple way of determining the wave length of the true waves which is equal to twice the distance between nodes.

Suppose a system of ropes is strung from a central point so they lie in a plane (see fig. 5). If the central point is given a regular up-and-down motion, waves will travel out along each rope and the system will present somewhat the appearance of still water into which a stone has been dropped. The wave trains traveling outward along each rope with the same velocity give the appearance of regularly growing or spreading concentric rings. The rings are separated by a distance equal to the length of the waves. If more ropes are added to this system so that they are stretched equally in every direction in various planes and the central point is given a regular to-and-fro motion the system will give the appearance of expanding or growing spherical shells. Here the distance between the shells is again equal to the length of the waves in the individual ropes. The mechanical rope apparatus is frequently used as an analogy to the electric field of a point charge.

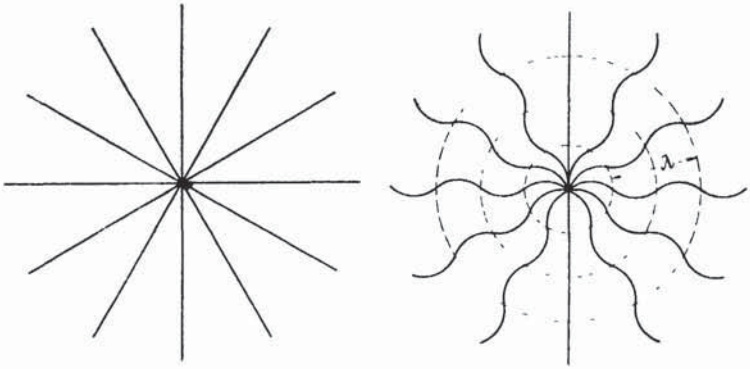

Figure 5. Radiation from a central point.
left: a rope system; right: waves in a rope system.

Definition of a Charge of Electricity. Electricity, according to present ideas, embodies two kinds called positive and negative. If an object has equal amounts of the two, it is said to be electrically neutral. If it has an excess of either kind it is said to be charged, and this excess is called an electric charge. Usually this charge is distributed over the surface of the object.

In discussions of the effects of one charge upon another, and in related problems, it is convenient to ignore the object and to think of the charge as being concentrated at one point. This is called a point charge.

The Electric Field of a Point Charge. We will now see why the three-dimensional system of ropes forms a rough mechanical analogy to the electric field of a point charge. Suppose a charge of positive electricity is fixed in space at some point A. If a charge of negative electricity is brought to some point B, it will experience a force which tends to draw it straight toward A as though the two were connected by an invisible stretched elastic cord. The point B may be anywhere in space in the vicinity of A and still it is drawn directly toward A. We may then think, if we like, of the space about A as filled or made up of these stretched elastic cords (called lines of force) extending outward in every direction from the charge at point A.

Now, if the charge at A were given a rapid to-and-fro motion, it would seem likely that waves should be formed and sped along the lines of force through space. This is the explanation offered by the wave theory of light concerning the manner in which radiant energy is propagated through space. We know that electric fields surround electric charges. Radiation is associated with waves traveling through these fields.

The waves in the electric field about the charge which has been set in oscillation are known to be not the only waves present. In fig. 3 is represented the shape of one of the lines of force shortly after the charge has been given a few oscillations. The train of waves which consists in variations in the direction of the line of force is traveling along this line with the velocity of light. Now it is known that the motion of an electric field through space produces an associated magnetic field. It therefore follows that this wave train must have associated with it a train of waves of magnetic intensity. These two wave trains lie in planes perpendicular to each other. The classical electromagnetic wave along one line of force is represented in fig. 6. Radiation is then nothing other than these electromagnetic waves.

The wave theory predicts that the velocity of propagation of these waves should be independent of their wave length. Moreover, such waves would be expected to exhibit all the phenomena of interference just as radiation does.

An oscillating charge radiates energy, of course, not only along one line of force but in all directions, though the amount of radiant energy sent out in different directions varies. Zero energy is radiated along the line of motion of the charge and the maximum amount is radiated in a plane normal to the direction of motion.

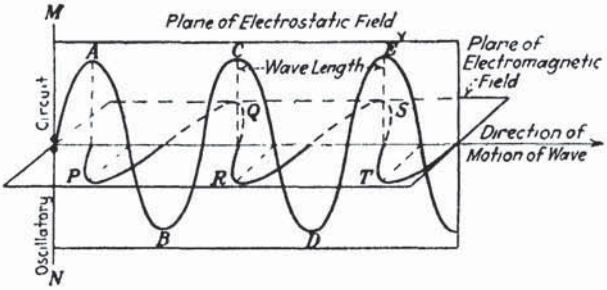
M
Plane of Electrostatic Field
Plane of Electromagnetic Field
A
C
E
Wave Length
Q
S
Direction of Motion of Wave
P
R
T
B
D
Oscillatory Circuit
N

Figure 6. Diagram of an electromagnetic wave.

Hertz (1866) caused a charge to oscillate rapidly between two closely-placed points[1]) and found that energy was being radiated as the result of the accelerations of the charge (see fig. 7). He placed a metal plane some distance from the oscillating charge

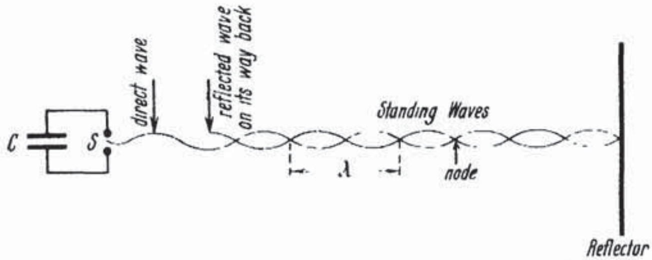
direct wave
reflected wave on its way back
Standing Waves
C
S
λ
node
Reflector

Figure 7. Experiments showing the wave nature of Hertzian waves.

---

[1]) In fig. 7, *c* represents a condenser, and *S* a spark gap. The condenser was charged to such a difference of potential that a spark occurred between the balls of the spark gap. Such an arrangement is known as an oscillatory circuit, for when a spark occurs the charge flows across the spark gap in such a way as to discharge the condenser, and it continues to flow until the condenser is recharged with the difference of potential reversed. The charge then begins to reverse its direction of flow. This process repeated many times a second amounts to a charge moving rapidly back and forth between the spheres of the gap or a charge oscillating rapidly between two closely-placed points.

and found regularly-spaced points between the radiating charge and the reflector at which the energy was alternately of a maximum and of a minimum value. These loops and nodes showed that the energy was being radiated in the form of transverse waves, the direct and the reflected beam interfering to cause standing waves.

When the velocity of these waves, which are of the wave length of short radio waves, was found to equal the experimentally-determined velocity of light, it was immediately suggested that light was nothing other than such waves, only of shorter wave length.

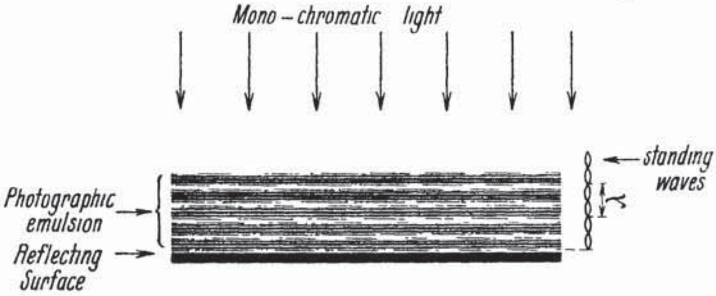

Figure 8. Experiment showing the wave nature of light.

A simple experiment performed by Lippmann showed that visible light could be made to form standing waves and thus must have a wavelike nature. Light of one wave length was allowed to shine perpendicularly upon a fine-grained photographic emulsion which was backed by a reflecting layer of metallic silver. When the emulsion was developed, cut and the edge viewed under a microscope (see fig. 8) alternate exposed and unexposed layers were found to exist throughout the depth of the emulsion At planes where the direct and reflected beams interfered to form the nodes of the standing waves, the silver compound was unaffected; at planes in between, corresponding to the loops of the standing waves, the silver compound was reduced. This experiment not only showed that light was a wave motion but offered a beautifully direct way of measuring the wave length.

Definition of Intensity. The intensity of radiation received from a source at a fixed point in space is defined as being the number of energy units (ergs) received per second by a square cm. of surface placed normal to the radiation at that point (see fig. 9a). When the radiation is strictly parallel, the intensity per cm^2 will be the same at any distance from the source (fig. 9a). With a point source, it will decrease as the second power of the distance between the radiating source and the point of measurement. Thus, in fig. 9b, A will receive K ergs per second while B, twice as far from the source, receives only $\frac{K}{4}$ ergs. In practice, point surfaces are rare, radiation generally being emitted by surfaces or volumes. (This is commonly the rule in biological radiations.) For these cases, the inverse square law holds only for distances so great that the source may be considered to be a point. For shorter distances the intensity may be roughly proportional to the reciprocal of the distance; for points still closer to the source, the intensity may be independent of the distance. Experiment is the best means of determining the variation of intensity with distance in the region of space closely surrounding a source of finite size.

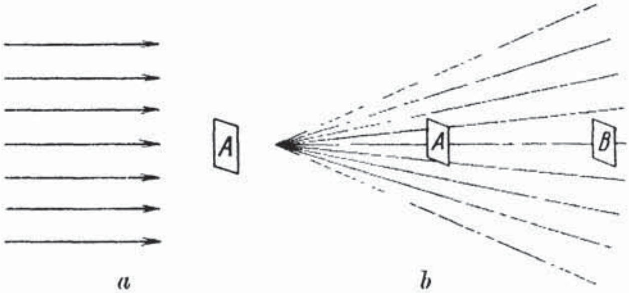

Figure 9. Illustration of the definition of intensity.

The wave theory of radiation has been successful in explaining how radiant energy may be transferred through space; it has predicted accurately the velocity of radiant energy; the phenomena of interference, reflection, refraction, dispersion, polarization and double refraction offer no difficulties; however, with respect to the emission and absorption of radiant energy, the classical theory fails and gives place to the quantum theory.

D. THE QUANTUM THEORY OF RADIATION

1. Definitions. It is known that matter exhibits the curious behavior of *discontinuity* in processes in which it emits or absorbs radiant energy. A given atom, for example, will

convert, of its store of energy, only certain multiples of the unit of energy into radiation. Likewise, it will absorb radiant energy only when the energy comes in precisely the proper-sized amounts. This phenomenon is one with which the wave theory of light is unable to cope.

Let us lay aside for the moment then this conception of the nature of radiation and consider the only other possible one, i. e. that radiation is corpuscular in nature. Thus we think now of radiation as consisting of small energy projectiles which travel through space with the familiar velocity of 3×10^{10} cm. per second. These projectiles are of course non-material; they consist simply of small units of energy.

Since we have seen that the different kinds of radiation, from radio waves to gamma rays, all travel with the same speed, these differences can occur only by differences in the size of each projectile, or quantum. It has been shown that the energy of a quantum can be given as the product of a universal constant, h, known as PLANCK's constant and equal to 6.55×10^{-27} erg seconds, and the frequency of the equivalent electromagnetic wave. Energy of quantum $E = h\nu$.

In Table 1, column I are given the various wave lengths in ÅNGSTROM units ($1\ \text{Å} = 10^{-10}$ m). Column II gives the corresponding frequencies obtained from the equation

$$\nu = \frac{c}{\lambda},$$

where c is the velocity of light. In the third column are the quantum energies, E, which correspond to each frequency ($E = h\nu$). Thus, an X-ray quantum is a 10^{-8} erg projectile, while a quantum of visible light has an energy value of but 10^{-12} ergs, and those of the ultraviolet being about 2 to 10 times as large as those of visible light.

For the understanding of emission and absorption of quanta by matter, it is necessary to discuss briefly the atomic theory.

2. Atomic Theory. Let us suppose (see fig. 10) that an electron is held at some distance, r, from a small positively-charged particle, q. From our knowledge of electrostatics we know that the electron experiences a force of attraction toward q. This force, F, is proportional to the number of units of charge possessed

Table 1. Photoelectric Energy Relations

	Wave Length in Å	Frequency	Energy of Quantum $E = h\nu$ (in ergs)	Electron Energy $V = \frac{12336}{\lambda}$ (in volts)
gamma rays	0.001	2.998×10^{21}	1.96×10^{-5}	1.23×10^{7}
	0.01	2.998×10^{20}	1.96×10^{-6}	1.23×10^{6}
x-rays	0.1	2.998×10^{19}	1.96×10^{-7}	1.23×10^{5}
	0.3	9.993×10^{18}	6.54×10^{-8}	4.11×10^{4}
	0.5	5.996×10^{18}	3.93×10^{-8}	2.47×10^{4}
	1.0	2.998×10^{18}	1.96×10^{-8}	1.23×10^{4}
	10.0	2.998×10^{17}	1.96×10^{-9}	1.23×10^{3}
ultraviolet	100.0	2.998×10^{16}	1.96×10^{-10}	1.23×10^{2}
	1000.0	2.998×10^{15}	1.96×10^{-11}	1.23×10^{1}
	1850 (Hg)	1.621×10^{15}	1.06×10^{-11}	6.67
	2000	1.499×10^{15}	9.81×10^{-12}	6.17
	2500	1.199×10^{15}	7.85×10^{-12}	4.93
	2536 (Hg)	1.182×10^{15}	7.74×10^{-12}	4.86
	2700 (Hg)	1.110×10^{15}	7.27×10^{-12}	4.57
	3342 (Hg)	8.971×10^{14}	5.87×10^{-12}	3.69
	3656 (Hg)	8.200×10^{14}	5.37×10^{-12}	3.37
visible light	4350 (Hg)	6.892×10^{14}	4.51×10^{-12}	2.84
	5000	5.996×10^{14}	3.93×10^{-12}	2.47
	5500	5.451×10^{14}	3.57×10^{-12}	2.24
	6000	4.997×10^{14}	3.27×10^{-12}	2.06
	6500	4.612×10^{14}	3.02×10^{-12}	1.90
	7000	4.283×10^{14}	2.80×10^{-12}	1.76
heat waves.	8000	3.747×10^{14}	2.45×10^{-12}	1.54
	10000	2.998×10^{14}	1.96×10^{-12}	1.23
	20000	1.499×10^{14}	0.98×10^{-12}	0.62
radio waves	10 cm to 600 m			

by q and is inversely proportional to the square of the distance, r, or

$$F = \frac{q}{r^2}.$$

Let us assume that q is heavy compared to the electron so that the electron will be the motile one of the two charges. If the electron is released from its position, it will, of course, fall toward and into the positively-charged body, q. Suppose, however, that the electron is set travelling in a circular orbit about q as the center (see fig. 11). If its orbital velocity is adjusted until its centripetal force is just equal to the force of attraction, F, exerted upon it by q, then the electron will be in stable equilibrium remaining indefinitely in this orbit.

q
r
electron

Figure 10.
Positive and negative charges at the distance r from each other.

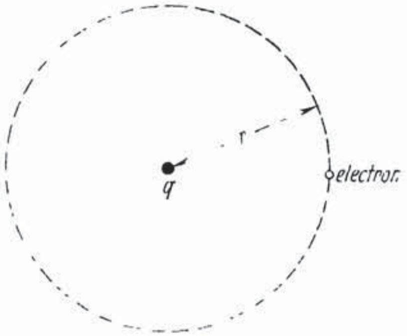

Figure 11.
Electron in a circular orbit traveling about the positive charge q.

An example of such a system is furnished by the sun and the earth. Here it is the balance between the earth's centripetal force due to its orbital velocity and the gravitational attraction of the sun which keeps the earth in its orbit.

If the mass of q is 1.65×10^{-24} grams, the distance r is 0.5×10^{-8} cm. and the charge q represents an electron, then fig. 11 represents a hydrogen atom. q, which is thus the unit of positive electricity, becomes the nucleus of the atom and the electron represents the hydrogen atom's one orbital electron.

From elementary considerations it would seem that the nucleus with its orbital electron would form a stable system also for values of r other than 0.5×10^{-8} cm. For any value of r, a corresponding orbital velocity can be calculated which will bring about the balance of the electrostatic attraction and the centripetal force of the electron. However, it is found experimentally that there are only a few orbits, out of the infinite number possible, which the orbital electron frequents. From the radius, r_1, of the innermost orbit of H which we have just seen to be equal to 0.5×10^{-8} cm. we may calculate the other possible orbits by the relation

$$r = n^2 r_1$$

where n has any value 1, 2, 3, 4 . . . . . .
The orbital velocity of the electron decreases as n increases, and the limiting case $n \rightarrow \infty$ corresponds to an atom with its electron at rest at an infinite distance from the nucleus.

The energy possessed by the atom or the nucleus-electron system, for the case when the electron is in any one particular orbit, can be easily calculated and this energy characterizes the *energy state* of the atom. The atom has the least energy when the electron is in the innermost orbit (state of least energy) and its energy increases as the electron exists in orbits of greater radius (states of higher energy). The innermost orbit is the preferred one, i. e. the electron inhabits this one the greater part of the time. In this case, the atom is said to be in its *normal* state. The usually unoccupied orbits are called *virtual orbits*. We have seen that the hydrogen atom can exist in different energy states, and we will very shortly apply this idea in a discussion of the absorption and emission of radiant energy by atoms (see pp. 14 and 16).

Before considering these topics, it will be advantageous to see how other atoms beside hydrogen are constituted. The one next in simplicity is helium. Its nucleus consists of 4 simple hydrogen nuclei, called protons, and 2 electrons. Two orbital electrons complete the atom. In all atoms, the number of protons in the nucleus exceeds the number of electrons by just the number of orbital electrons; this leaves the nucleus with a positive charge and the whole atom electrically neutral. For example, the carbon atom is composed of a nucleus of 12 protons and 6 electrons about which revolve 6 electrons in the various shells. The oxygen atom consists of 8 outer electrons and a nucleus which contains 16 protons and 8 electrons. The atomic weight of carbon is approximately 12 and that of oxygen is 16. It can be seen that the atomic weight is equal, for all practical purposes, to the number of protons in the nucleus of the atom. The atomic number is equal either to the number of nuclear or orbital electrons. Table 2 shows in what way the outer electrons group themselves in the orbital shells for the first eleven elements of the periodic table.

Table 2. Electron Configuration for the Elements from H to Na.

Element	Atomic Number	*K* shell	*L* shell		*M* shell
H	1	1			
He	2	2			
Li	3	2	1		
Be	4	2	2		
B	5	2	2	1	
C	6	2	2	2	
N	7	2	2	3	
O	8	2	2	4	
F	9	2	2	5	
Ne	10	2	2	6	
Na	11	2	2	6	1

Fig. 12 is a wholly diagrammatic representation of the *Na* atom. The central dot marks the nucleus; the first circle is called the *K* shell and contains two electrons. The binding energy of these negative charges is about 1000 electron volts or 1.6×10^{-9} ergs; this amount of energy would be required to remove either one of the *K* electrons from the atom. The next circle representsthe *L* orbit, which in *Na* contains 8 electrons, the binding energies of which are approximately 35 electron volts. In the last solid circle there is but one electron and its binding energy is about 5 electron volts. The outer dotted circles represent energy levels which are unoccupied in the normal *Na* atom, i. e. virtual orbits.

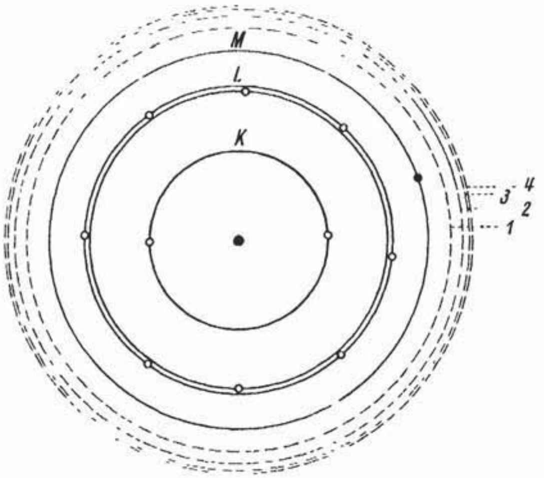

Figure 12. Diagram of the sodium atom showing the various shells.

3. The Emission and Absorption of Radiation by Atoms. We have seen that the hydrogen atom can exist in various energy states associated with the orbit occupied by its electron.

By this necessary mechanism can be explained the absorption and emission of radiation. If the atom changes from an energy state E_m to a lower one E_n, it does so with the emission of a quantum of radiant energy $h\nu$ such that

$$h\nu = E_m - E_n. \tag{1}$$

If the atom changes from an energy state E_n to a higher state of energy E_m, it can do so only by the absorption of a quantum of radiation $h\nu$ of such energy value that

$$h\nu = E_m - E_n. \tag{1}$$

In this equation h is the universal constant known as PLANCK'S constant equalling 6.547×10^{-27} erg. secs. and ν is the frequency (see p. 4). It follows that the various quanta $h\nu$ may be emitted and absorbed by the hydrogen atom.

$$\begin{array}{llll} h\nu = E_m - E_0 & \text{where} & m = 1, 2, 3 \ldots\ldots \\ h\nu = E_m - E_1 & ,, & m = 2, 3, 4 \ldots\ldots \\ h\nu = E_m - E_2 & ,, & m = 3, 4, 5 \ldots\ldots \\ h\nu = E_{n+1} - E_n. & & \end{array}$$

Since $\lambda = \frac{c}{\nu}$, where c is a constant which is equal to the speed of light or 2.99796×10^{10} cm. per sec., the wave length of the radiation resulting from or producing the energy change $E_m - E_n$ in the atom will be given by the equation

$$\lambda = \frac{ch}{E_m - E_n}$$

Table 3 gives the values of the first eight of the forty or so known energy states of the hydrogen atom.

Table 3. Energy obtained by electron shifts from normal to higher orbits in the Hydrogen atom

number of virtual orbit	E in ergs 10^{-12}	corresponding wave length in Å
0	0	0
1	16.14	1216
2	19.15	1025
3	20.18	975
4	20.70	948
5	20.90	940
6	21.05	933
7	21.16	928

The wave length emitted by the hydrogen atom for the energy change $E_1—E_0$ for example, is readily calculated from the data in this table, as

$$\lambda = \frac{2.99796 \times 10^{10} \times 6.547 \times 10^{-27}}{161.4 \times 10^{-13} — 0} =$$

$$1216 \times 10^{-8} \text{ cm. or } \lambda = 1216 \text{ Å.}$$

This wave length is in the far ultraviolet. Changes between other states result in the radiation of visible light and still others produce radiation in the far infra red, e. g. for the shift from the 6th to the 7th orbit, it is 178 500 Å.

The equation (1) states that an atom in the state E_n will absorb a quantum $h\nu$ and be raised to the energy state E_m if the quantum is precisely equal to the difference in the energy of the two states. Supposing the energy of the quantum is slightly less than this difference—will it be absorbed? The answer is no; there is no possibility that it will be. If the quantum is larger than $E_m—E_n$, then it may be absorbed. It will be, of course, if its energy happens to equal the energy difference between any two states provided that, at this moment, the electron is in the orbit corresponding to the lower of these states. If its energy is not one of these discrete values, it will not be absorbed — *unless* its energy is greater than $(E_\infty — E_n)$, i. e. sufficient to shift the electron beyond the outermost orbit. In this event, it may be absorbed and the surplus, $h\nu — (E_\infty — E_n)$, is used in shooting the electron away from the atom, or

$$h\nu = E_\infty — E_n + \frac{1}{2} mv^2$$

where m is the mass of the electron and v is its velocity (the term $\frac{1}{2} mv^2$ represents the kinetic energy of the ejected electron). An electron so ejected from an atom is called a *photoelectron*, and the atom itself is said to be *ionized*.

As has been stated before, an electron spends most of its existence in the normal state, E_0. When it has been raised to a state of higher energy E_n by virtue of the absorption of energy, the atom is said to be in an *excited* state. The life of an atom in an excited state is of the order of 10^{-7} to 10^{-9} seconds. After this length of time the atom reverts to its normal state with the resulting emission of radiant energy.

These remarks on emission and absorption of radiation apply not only to hydrogen but to the other atoms as well. Only the outer electrons of the more complicated atoms behave in a manner similar to the one electron of hydrogen. In this way originate the atomic spectra. They are emitted by sources (such as the mercury arc or a glow discharge tube) in which the gas is at sufficiently low pressure that the atoms are not in contact with each other but for a small fraction of the time. If the pressure is raised (also the case for solids), a *continuous* spectrum is emitted which does not contain lines characteristic of the atom. This is called *thermal* radiation since it is the result of the temperature of the source. The atoms are so close together that the outer virtual orbits intermingle and are distorted.

These last remarks apply equally well to absorption. An element in the gaseous state will give a line absorption spectrum, while in the solid state it will give continous absorption.

Molecular Spectra

While atomic radiation results from electrons jumping from one energy level to another, molecular spectra are assumed to arise from the motions of atoms or, better, ions which form the molecule.

Let fig. 13 represent a simple diatomic molecule such as NO. Such a molecule emits radiation in three wave length regions: (1) the far infra red; (2) the near infra red; and (3) the visible or the ultraviolet regions. The radiation in the first group is ascribed, in the simple theory, to changes in the rotational energy of the dipole molecule. The second group is ascribed to simultaneous changes of rotational and vibrational energy, and the third, to simultaneous changes in rotational, vibrational and electronic energy of the molecule.

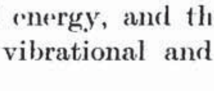

Figure 13.
A simple diatomic molecule.

Molecular spectra are in general exceedingly more complex than atomic spectra. The great number of lines fall into groups which under low dispersion give the appearance of bands.

The absorption spectrum of a molecule is, of course, its emission spectrum in reverse, light bands replaced by dark.

In this connection, the effect of radiation upon chemical reactions should be mentioned. To realize that such an effect does exist, it is only necessary to remember the number of brown bottles that are used for storing chemicals. The most prominent effect is that of light on silver salts as in all photographic emulsions. The result is the reduction of the salt with the deposition of metallic silver. Without presenting the theory of the process, it is still easy to see that the absorption of radiant energy might do just this. For an absorption of energy means that the molecule must go into a state of higher energy-a less stable state- and this may, on the absorption of sufficient energy, become a chemically unstable state.

E. ANALYSIS OF RADIATION BY DISPERSION INTO A SPECTRUM

The radiation emitted by a source is characterized by the wave lengths present, together with the distribution of energy among these wave lengths. A given atom will emit only certain lines (its line spectrum) and each one will have associated with it a certain energy (see upper spectrum of fig. 14); a molecule may give rise to some such spectrum as that of the center strip, and an incandescent solid emits all wave lengths with varying intensity beyond a certain wave length, depending upon the temperature of the solid (see lower spectrum of fig. 14).

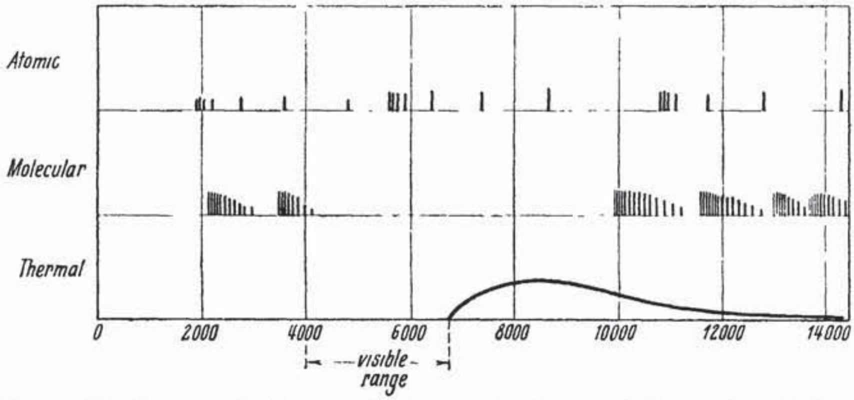

Figure 14. Representation of atomic, molecular, and thermal radiation.

The various wave lengths present in the radiation emitted by a source are determined by dispersing the radiation into a spectrum. This subchapter deals with the instruments and methods used to produce a radiation spectrum both in the visible and the ultraviolet. The next deals with the subject of measuring the intensity associated with the various wave lengths.

The two instruments most frequently used to disperse light into a spectrum are the prism and the grating. When a narrow beam of parallel light falls upon a prism, the different wave lengths present in the beam suffer different deviations in passing through the prism. As a result, each wave length emerges with a slight angular separation from its neighbors. Since, in practice, beams of light are generally divergent rather than parallel, the prism alone gives rise to a spectrum in which there is some overlapping of the wave lengths.

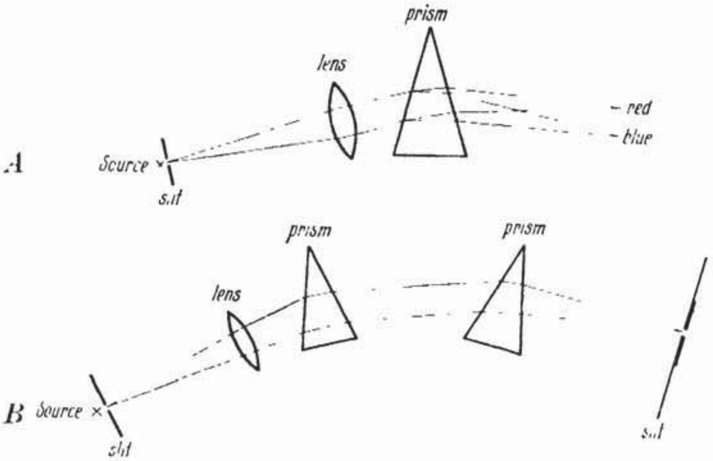

Figure 15. Spectrometers
above: a simple spectrometer; below: a quartz double monochromator.

The simple optical spectrometer (see fig. 15A) prevents this by employing a lens which forms in the eyepiece a narrow image of the slit through which the light enters. By replacing the eye piece by a slit, the instrument may be used as a monochromator or monochromatic illuminator.

For work in the ultraviolet region, the prisms and lens must be of quartz. Fig. 15B represents the optical system of a typical quartz double monochromator. In most instruments the prisms may be rotated by some device which is connected to a drum calibrated in wave lengths. Turning this drum to a certain wave length reading sets the prisms so that only this wave length is permitted to pass through the second slit. Table 4 gives the relative intensities of the lines in the ultraviolet spectrum of the quartz mercury arc as transmitted by such a monochromator.[1]

Table 4. Relative Intensities of Hg Spectral Lines

Wave Length Å	Relative Intensity	Wave Length Å	Relative Intensity
2967	528	2345	20.0
2925	117	2302	10.6
2894	209	2284	13.6
2804	371	2253	6.6
2754	63	2225	7.9
2700	88.6	2191	4.75
2653	735	2150	2.74
2536	1000	1973	0.128
2482	218	1943	0.097
2399	51.5	1850	0.017
2378	40.0		

Gratings are also used to produce spectra of visible and ultraviolet light but since their application is more strictly confined to special work in spectroscopy, a discussion of their characteristics will be omitted.

While monochromators are designed to give high spectral purity of the isolated light, some degree of impurity seems unavoidable. In general, some intensity of radiation of shorter wave length than that desired is transmitted by the instrument. This defect may be greatly reduced by using with the monochromator a filter having a "cut off" on the short wave length side of the desired radiation. By so doing, the intensity of the unwanted wave lengths may be reduced to practically zero. An excellent list of such filters is given in a table (12—5) in *Photoelectric Phenomena* by Hughes and Du Bridge and the ultraviolet portion is reproduced here with their kind permission (see Table 5). The wave lengths given in the table are those at which the filter ceases

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[1] In particular, a Hilger quartz double monochromator.

Table 5. Short Wave Cut-off Filters

Cut-off	Material	Thickness, etc.	Comments
900 A	thin celluloid	30 to 40 mμ	Gradual cut-off
1230 A	clear fluorite	1 to 2 mm	Only very occasional specimens transmit as far as this
1450 A	clear quartz (crystalline)	0.2 mm	Different specimens have practically identical transmission limits
1500 A	clear quartz (crystalline)	2 mm	Air paths have strong absorption below this point
1600 A	clear quartz (crystalline)	20 mm	
1700 A	oxygen in a quartz (crystalline) cell	10 mm, at atmos. pressure	
1750 A	water in a quartz (crystalline) cell	20 mm	Steep cut-off. (Lyman finds, however, that 0.5 mm of water has a sharp cut-off at 1729 A)
1750 A	clear fused quartz	0.3 mm	Transmissions: 1849 A, 24% 1971 A, 36% 2002 A, 40%
1850 A	quartz mercury lamp		1850 A is the shortest wave length emitted by a new lamp
2000 A	clear fused quartz	3 mm	Transmissions: 2000 A, 0% 2100 A, 56% (different specimens vary widely in transmission)
1900 A	acetic acid in water	32 mm, 1 part acid in 1000 parts water	The concentrations are merely rough estimates; it is best to find by trial the desired concentration
2000 A	acetic acid in water	32 mm, 1 part acid in 200 parts water	
2200 A	code 970[1])	5 mm	Gradual cut-off; "solarizes" with short wave lengths
2200 A	calcite	10 mm	
2300 A	rock salt	12 mm	Lyman and Pfluger, however, found rock salt to transmit as far as 1750 A

[1]) standard filters of the Corning Glass Works.

Cut-off	Material	Thickness, etc.	Comments
2350 A	p-dichlorbenzene	50 mm; sat. solut. in water	
2450 A	tartaric acid	3.2 mm; 1 part sat. solut. in 64 parts water	
2500 A	thiophene	50 mm; sat. solut. in water	Steep cut-off
2800 A	benzol		Steep cut-off
2800 A	code 971 1)	2.1 mm	Fairly steep cut-off
2900 A	code 971 1)	4.9 mm	Fairly steep cut-off
3000 A	pyrex 1)	1.0 mm	

to transmit appreciably. These authors point out that "a filter is seldom found in which the transmission changes from 50 per cent to 1 percent or less in 200 Å".

In some cases it is possible to find a source giving widely separated lines in a desired wave length region. By using a suitable filter with such a source, it frequently occurs that but one line is transmitted. Such a combination may give much greater intensity than could be obtained by the use of the monochromator.

Because of the great intensity of solar radiation and the possible effects of daylight on biological reactions, some data are given on the short wave length limit of the solar spectrum. This limit of the solar spectrum as recorded by a photographic plate at various altitudes of from 50 to 4560 meters was found by one observer to be 2910 Å. Another experiment carried out at 9000 meters showed energy present at 2897 Å. The conclusion is given that the intensity, at the surface of the earth, of the solar radiation of wave length 2900 Å is not more than one-millionth of the intensity at 3150 Å. This sharp cut-off is ascribed to the ozone present in the upper layers of the atmosphere. Nevertheless, though the absorption approaches asymptotically 100%, we must keep in mind the possibility that extremely small intensities of the lower wavelengths exist in day light. This is important since in biological radiations, we are dealing with very low intensities, far beyond detection by photographic plates.

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1) standard filters of the Corning Glass Works.

Table 6 gives the absorption of ultraviolet by 16.97 mm of distilled water. This, together with the absorption spectrum of oxygen, shows plainly why in biological radiations, no experiments are carried below 1900 Å.

Table 6. The Absorption of Ultraviolet in Distilled Water (16.97 mm)

Wave Length Å	Absorption %
1860	68.9
1930	24.5
2000	14.2
2100	9.8
2200	9.2
2300	5.6
2400	5.2
2600	4.2
3000	2.5

F. THE INTENSITY MEASUREMENT OF VISIBLE AND ULTRAVIOLET RADIATION

Arranged in order of increasing sensitivity, the detectors of visible and ultraviolet light are: the thermocouple, the photographic plate, the photoelectric cell and the photoelectric counter.

(1) The Thermocouple: The thermocouple consists of a circuit composed of two dissimilar metals or alloys (see fig. 16a).

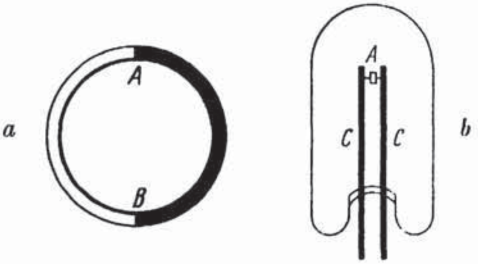

Figure 16. Thermocouples left: a two-metal thermo-element; right: a modern high-sensitivity thermocouple.

If the junctions are maintained at different temperatures, a current will flow in the circuit, which is proportional to the temperature difference of the two junctions. Modern high-sensitivity thermopiles are frequently built as diagrammed in fig. 16b in which *A* is a very light bit of thin gold leaf, 1 or 2 mm² in area. The gold leaf is supported by two fine wires, one of a bismuth-antimony alloy, the other of bismuth-tin, which are soldered to the heavy leads, *C*. By using pieces of small dimensions, the heat capacity of the instrument is low, as is the heat loss by conduction: thus, the sensitivity is high. Used with a sensitive galvanometer, such a thermopile will give a detectable deflection when the radiation falling upon the gold leaf has an intensity of approximately 3×10^{-10} cal/cm²/sec. or 1×10^{-2} ergs/cm²/sec. The advantage possessed by the thermopile over the other means of intensity measurement is that its response is quite independent of the wave length of the radiation. For this reason, it is often used to calibrate a source, of ultraviolet for instance, in terms of the energy in the form of visible light, emitted by a standard tungsten lamp. The disadvantage of the thermopile is its low sensitivity as compared to the other instruments for this type of measurement.

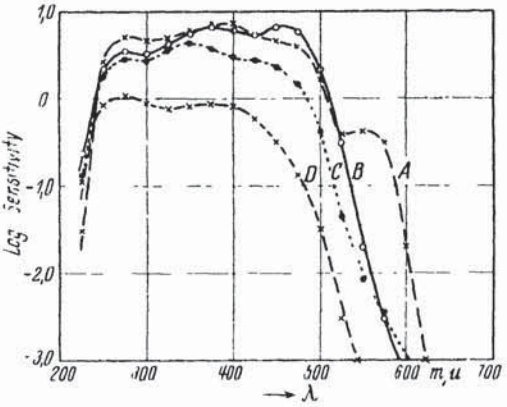

Figure 17. Spectral sensitivity curves of Eastman plates
A: Eastman Speedway; *B*: Eastman 40; *C*: Eastman 35; *D*: Eastman Process.

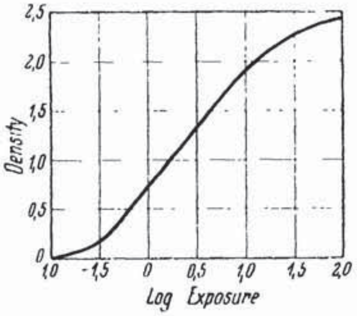

Figure 18.
Characteristic curve of a photographic plate.

(2) **The Photographic Plate.** Photographic plates manifest the defect just mentioned, namely that of giving a response which is *not* independent of wave length (see fig. 17 taken from C. E. K. MEES, 1931). Furthermore, the blackening of the emulsion for one wave length is proportional to the intensity only over a relatively short intensity range, perhaps 1 to 20 (see fig. 18). A paper rich in information on the characteristics of photographic emulsions is that of L. A. JONES and V. C. HALL (1926).

Plates have been evolved which are particularly sensitive to different wave length regions; the SCHUMANN plate, for example, in the ultraviolet. The sensitivity of the SCHUMANN plate for the ultraviolet depends upon the absence of gelatin. Gelatin has a strong absorption for wave lengths below 2800 Å and becomes practically opaque even in very thin layers for wave lengths in the neighborhood of 2000 Å.

Recently, even better plates have been obtained by sensitizing ordinary plates. They are covered with a very thin film of a substance (a carboxylic ester of dihydro-colloidin — furniture polish) which fluoresces strongly under the action of ultraviolet with the emission of longer wave lengths which easily penetrate the gelatin. About the lowest sensitivity to which the photographic emulsion will respond is 12000 quanta/cm^2/sec, or 10^{-7} ergs/cm^2/sec. at a wave length of 2500 Å.

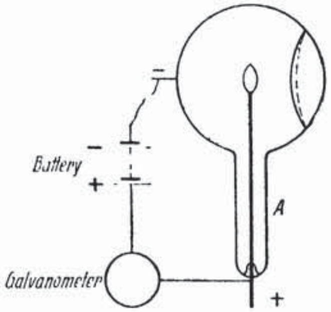

Figure 19.
Photoelectric cell.

(3) The Photoelectric Cell. In photoelectric cells, radiation falls upon a metal surface; the photoelectrons (see p. 16) ejected form a photoelectric current which is proportional to the intensity of the radiation. A linear relationship between the photoelectric current and intensity has been tested in properly-designed cells and found to hold over a range of intensities of from 1 to 50 million. This is many times greater than the linear portion of the photographic emulsion curve which extends over an intensity range from 1 to 20. Not all commercial cells give this behavior, however. In fact, it is quite unsafe to assume a linear relation between photoelectric current and light intensity for any but specially-made cells. The sensitivity of photoelectric cells is roughly that of the photographic plate.

Photoelectric cells are of two kinds, vacuum and gas-filled. In principle they operate in the same way. A glass bulb (see fig. 19) has deposited over most of its inner surface a coating of the photoelectric element, e. g. Na, K or Mg. This coating is in contact with a wire which is sealed through the glass wall of the bulb. Another wire is sealed in a side arm, *A*; a battery maintains the coating negative with respect to the wire at *A*. With the window covered so that no light can enter the cell, the galvanometer reads zero indicating that no current is flowing in the circuit. If light is allowed to fall upon the metal surface, photoelectrons are ejected from the atoms of the metal film and are attracted to the central wire. These electrons flowing through the wire cause the galvanometer to deflect, which is a measure of the photoelectric current.

Necessarily, in such cells, the energy, $h\nu$, of each quantum to be measured must be greater than the energy required to remove an electron from the surface, that is $h\nu > E\infty - E_0$. This equation applies to isolated atoms. In a photoelectric cell, the photoelectrons must not only be removed from the atoms but must be shot away from the metal surface and eventually even through it. This requires a little more energy. The energy required to remove a photoelectron from a photoelectric surface is called the "work function", W_0.

The situation in the sensitive surface of a photoelectric cell is further complicated because of the impossibility of having this surface consist of one kind of atom. With the best high vacuum technique known today it is impossible to prevent contamination with various atoms, chiefly those of the gases prevalent in the air, such as oxygen, hydrogen and nitrogen. The result is that W_0 for a given metal depends considerably upon its history and the care with which it has been freed from gases. The purest surfaces are prepared by distilling metals in a high vacuum. Table 7 gives the photoelectric work functions, W_0, for various metals as obtained by different investigators; only a few metals being given, since these may be regarded as representative. (Further data may be found in *Photoelectric Phenomena*, HUGHES and DU BRIDGE). The threshold wave length in Å refers to the longest wave length which will eject photoelectrons from a given surface, and this may be transformed into a value in electron volts—then called the photoelectric work function — by the equation

$$W = \frac{12336}{\lambda \text{ in Å}}.$$

The first three columns show how the value of the long wave length limit or the work function depends upon the treatment given the surface. It is rare in this work to find the results of two investigators coming within more than approximate agreement.

Table 7. Photoelectric Work Functions of the Metals

Metal	Threshold Wave Length in Å			W_0 in volts
	no outgassing	partial outgassing	extended outgassing	
Ag	3213	2888	2610 (20°C)	4.73
	3250	3150	2700 (600°C)	
	3364			
	3390			
Al	3460	3652		
	3595			(2.5 to 3.6)
	3650			
	4132			
	4770			
	5000			
Cd	3050			(4.00)
	3130			
	3140			
	3302			
K	~4360	5500	5500	(1.76 to 2.25)
	6700	5800		
		5800		
		6200		
		6500		
		7000		
		7000		
Li	5260	4300	5400	(2.1 to 2.9)
		5200		
		5600		
		5800		
Mg	3300	≥3650		(~3.40)
	3820			
	7000			
Na	5830	5500	5000	1.90 to 2.46
	6100	5500		
		6400		
Ni	3050	2700	2463	5.01
	3365	3040		
Zn	3016	3182	3720	3.32
	3200		3460	3.57
	3425		(Single	
	3760		crystal)	
	4009			

The fourth column gives the best estimate of the value of W_0 that could be made for the various metals.

(4) **The Photoelectric Counter.** The photoelectric tube counter is merely a photoelectric cell of a special geometrical shape. In such an instrument, the individual photoelectrons are recorded, making it much more sensitive than the photo cell in which photoelectron currents are measured. The lowest intensity which is measurable with a counter is about 10^{-9} ergs/cm^2 sec. or about 500 quanta/cm^2/sec.

In a counter, the photoelectrically active element is deposited on the inside walls of a cylinder, and the collector is a fine wire insulated from the cylinder and stretched along its axis (see fig. 20). It is filled with some gas to a pressure of about 10 cm of mercury. The metal tube is connected to the negative terminal of a battery of perhaps 1000 or 1500 volts, and the collecting wire to an amplifier, such as is found in a radio receiver. Slits may be cut in the cylinder to let in the light, or it may be allowed to shine in the ends of the cylinder. Each time a photoelectron is ejected from the walls of the tube, it will be accelerated toward the wire (which is positive by 1000 or 1500 volts) and in its course through the gas will ionize some of the atoms with which it collides. The electrons thus freed are also attracted toward the wire and in turn form more ions. In a very small fraction of a second all these negative ions will reach the wire. This momentary movement of charge is equivalent to a small current which when amplified, produces a "plunk" in the loud speaker. The number of "plunks" per second indicates the number of photoelectrons ejected per second; this number is proportional to the number of quanta striking the inner wall of the tube each second. While in use, the counter gives a few counts per minute when no radiation from the source under experiment is falling upon it. These are due to cosmic radiation, local gamma radiation and α, β and γ rays from radioactive impurities in the metal of the wire and tube; they are

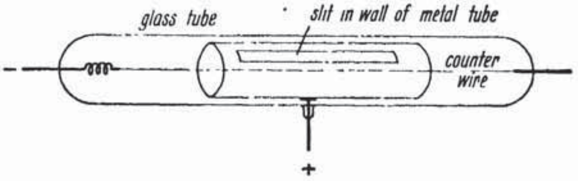

Figure 20. Photoelectric tube counter.

called "dark counts", "strays" or background radiation. The difference between the number of counts when the counter is exposed to and shielded from radiation is proportional to the intensity of the incident radiation. Fig. 21 illustrates data taken with an aluminum counter by **Frank** and **Rodionow** (1931) to prove the radiation from chemical reactions and from tetanized muscle The dotted line indicates the number of "strays" and the solid line the total number of counts when the counter is exposed to the radiation, a 5 minute exposure being alternated with 5 minutes of shielding.

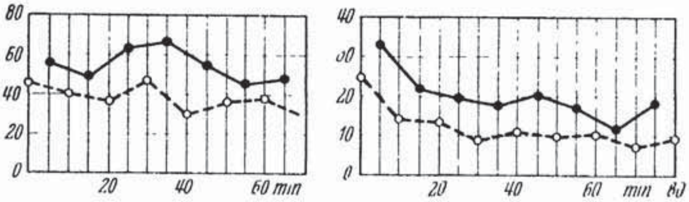

Figure 21. The ordinates are the number of impacts obtained per 5 minute interval, when the counter is exposed alternately for 5 minutes and shielded from the source of radiation. The source of radiation was at the left the chemical reaction $K_2Cr_2O_7 + FeSO_4$, at the right a tetanized sartorius muscle of the frog.

It has been found that for the ordinary counter (as well as for the photoelectric cell) only about 1 quantum in 10000 striking the walls ejects a photoelectron into the gas of the tube The number of quanta incident upon a surface divided by the number of photoelectrons ejected, or the average number of quanta required to eject one photoelectron, is called the *photoelectric yield* of the surface. The photoelectric yield is the reciprocal of the efficiency of a surface. A perfectly efficient surface would yield one photoelectron for every incident quantum.

Yields of surfaces in photoelectric counters have never been as high as those in photoelectric cells because the active gases H, O, etc. with which these counters are filled will reduce the sensitivity of the surface. It should be possible to fill a counter having a highly sensitive surface with some inert gas which will not reduce the sensitivity. **Werner** (1935) recommends 75 % Ne and 25 % He. However, the experience of one of the authors shows that it is difficult to obtain sharply defined counts with this mixture. More literature is quoted in Chapter IV, p. 91.

The values of Table 8 represent maximum yields obtained by experienced workers. Generally, yields of 100 or even 1000 times these values are considered to be good.

Table 8. Highest yields obtained with various surfaces at the maxima of their spectral distribution curve

Metal	Wave length in Å	Yield expressed in coulombs per cal.	Yield expressed in quanta per electron
Al	2360	0.01×10^{-2}	763
K	4200	3.00×10^{-2}	28
Mg	2500	5.20×10^{-2}	16
Na	3400	0.57×10^{-2}	134

CHAPTER II

SOURCES OF RADIANT ENERGY

Radiant energy may originate under widely varying conditions. Warm bodies radiate heat. When the temperature rises very high, the radiation becomes visible. Practically all our sources of illumination, from the oil lamp to the incandescent light are based on this principle. However, visible radiation may also be produced from chemical processes, without great increases in temperature, as in the slow oxidation of phosphorus or in the light of the firefly. Too, when an electric current is set up in a tube containing gas at low pressure, though the temperature remains within a few degrees of the room, the tube will emit light.

In this chapter, a division is made between physical and chemical sources of radiation. Though the chemical sources of radiation are more important because they show us that we may expect radiations in biochemical processes, the physical sources are much better known. The discussion begins therefore with the physical sources of rays, followed by the chemical sources of such rays; in Chapter III, the biological effects will also be first demonstrated with rays of physical origin, and afterwards with those emitted by chemical reactions.

A. PHYSICAL SOURCES

Thermal Radiation: Returning for the moment to the wave theory of light, we remember that the oscillations of an electric charge result in the production of radiant energy. Let us see how this idea may be applied to the various sources of radiation with which we are familiar. There is, first, the fact that all bodies emit at all times radiant energy; they also absorb at all times radiant energy. This radiation is due to the vibrations of the atoms (built of electric charges) of which the body is composed. At ordinary temperatures, this radiation is of very long wave length. However, the radiation becomes visible when the temperature of the body reaches the neighborhood of 500° C. — witness the dull red color of iron which is being heated. At still higher temperatures, e. g. that of the tungsten filament in an electric bulb, the color of the radiation is changed to nearly white, and in some of the hotter stars to a blue-white color. Radiation which arises as a result of the temperature of a body is known as *black body* or *thermal radiation*. The peculiarity of a thermal radiator is that the distribution of energy among the wavelengths depends not at all upon the atoms or molecules of the body, but solely upon its temperature. Table 9 gives the energy radiated by a tungsten filament at various wave lengths for the two temperatures 2500° and 3000° C. It can be seen from these data that thermal radiators are poor sources of ultraviolet.

Table 9. Spectral Distribution of Thermal Radiation from Tungsten

Wave Length in Å	$F_w(\lambda)$ (watt/cm^3) 1)	
	2500° C	3000° C
2000	0.07	8.32
2500	7	315
3000	125	2980
3500	869	13100
4000	3360	35900
4500	8930	73100
5000	18400	122000
5500	31400	176000
6000	47600	230000
6500	65200	276000
7000	82700	315000

Atomic Radiation: Everyone is familiar with the neon signs so frequently used at present for advertising purposes. These are nothing more than discharge tubes filled with neon or a mixture of neon and other gases. They are fitted with metal

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1) $E_w(\lambda)$ is the rate of emission of energy in watts, from 1 cm^2 of surface, in a direction perpendicular to the surface, per unit solid angle, for 1 cm range of wave lengths.

terminals to which electrical potentials are applied, potentials high enough to cause ionization of the gas atoms in the tube. The atoms lose and regain electrons many times a second with the emission of light each time an electron is regained. Such tubes are known variously as arc, glow and discharge tubes depending upon the pressure of gas within them and the voltage necessary to make them function. The most practical sources of ultraviolet, namely the mercury arc and the hydrogen arc are of this type. Other sources may depend simply upon the ionization of air between two naked terminals, such as the carbon arc or the iron or tungsten spark. The latter two are strong sources of ultraviolet but suffer from the disadvantage that they are not sources of constant intensity. In this type of source the temperature is not necessarily much different from room temperature — it is the electrical energy which causes the ionization of the atoms and the resultant emission of light.

B. CHEMICAL SOURCES

Most of the chemical reactions which proceed spontaneously are exothermic, i. e. they liberate energy. Ordinarily, the energy is emitted in form of heat, as the name "exothermic" implies. Occasionally, however, the reaction causes luminescence, the energy being liberated as visible light. As examples may serve the light produced during the slow oxidation of phosphorus, at the hydrogen-oxygen combination, at the reaction of potassium with water, or at the oxidation of pyrogallic acid. Haber has shown that these are not cases of light produced by heat, but that part of the original energy of reaction is liberated in the form of visible light.

By far more common is the emanation of ultraviolet light Recent investigations make it appear very probable that all chemical reactions emit part of their energy in the form of short ultraviolet rays. This has been proven for such simple processes as $NaOH + HCl = NaCl + H_2O$, and even for the solution of NaCl in water, i. e. $NaCl = Na^+ + Cl^-$, as will be shown later.

Ordinarily, the radiations are very weak, altogether too weak to be registered by the photographic plate. However, it has been possible to prove their existence by the Geiger-Müller counter which is essentially an extremely sensitive photoelectric cell (see fig. 21).

In this way, **Frank** and **Rodionow** (1932) proved that a number of common chemical oxidative reactions produced an ultra-violet radiation which could be demonstrated with a sufficiently sensitive instrument. Table 10 gives their results. It was also observed at this time that in some of the reactions, the emanation is greatly increased in the presence of diffuse day light. This physical proof of light from chemical reactions has been verified by **Gerlach** (1933) who showed that this radiation appears only from quartz vessels, not from glass containers; therefore, it must be of a wave length shorter than 3500 Å. **Audubert** and **van Doormal** (1933) also measured photo-electrically the emission of ultra-violet light by several inorganic oxidations, by the oxidation of alcohol with chromic acid, and by pyrogallic acid in air. Recently, **Barth** (1934) could also prove the existence of ultra-violet emission from proteolysis which is known to give an immeasurably small heat of reaction. In all of 25 experiments but one, the number of photo-electrons was larger when exposed to radiation from proteolysis than without this, and in 7 experiments it was more than 3 times as large as the error.

Table 10. Ultra-violet radiation from chemical reactions

Chemical Reaction	Time Interval	Photon impacts per time interval		Increase in impacts by chemical reaction	
	minutes	exposed	control	absolute	percents
Pyrogallic acid + NaOH + air	10	15 ± 1.2	10 ± 1.0	5	50
	8	38 ± 2.0	26 ± 1.8	12	46
$SnCl_2 + HgCl_2$	10	29.5 ± 1.7	26 ± 1.6	3.5	13
$FeSO_4 + K_2Cr_2O_7$ (dift. light)	6[1]	18 ± 1.7	13 ± 1.5	5	38
,, ,,	4[1]	17.5 ± 2.1	13 ± 1.8	4.5	34
,, ,,	6[1]	18 ± 1.7	12 ± 1.4	6	50
,, ,,	9[1]	36 ± 2.0	26.5 ± 1.7	9.5	36
$FeSO_4 + K_2Cr_2O_7$ (dark) . .	15	27 ± 1.3	17 ± 1.0	10	60
,, . .	4[1]	12 ± 1.8	11 ± 1.7	1	9

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[1] Radiation passed a monochromator, only the rays between 2000 and 2700 Å were measured.

These radiations are so very weak that only the most sensitive counters will detect them. However, living cells under certain physiological conditions react very promptly upon irradiation in the wave length range 1800—2600 Å. In fact, they are so sensitive that it has been possible to use them in place of photographic plates for determining the spectra of such radiations. The organisms most used in these experiments are yeasts the growth rate of which is accelerated by short ultraviolet rays under certain conditions. The biological aspects of this acceleration will be discussed in Chapter IV.

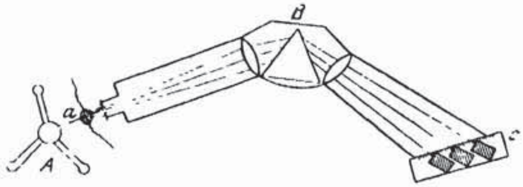

Figure 22. First attempt to obtain a mitogenetic spectrum

a: an electrically excited muscle: *B*: the quartz prism; *C*: agar blocks with yeast on the exposed side, each block receiving rays of known wave length.

The original method consisted simply in placing before the collimator slit of a quartz spectrograph a quartz tube with the reagents to be tested, and to substitute the photographic plate by a succession of tiny blocks of nutrient agar on which yeast in the proper physiological condition was growing. Fig. 22 shows the first attempt, by FRANK (1929) to obtain the spectrum of a frog muscle. Each yeast block was thus exposed to a definite range of the spectrum which could be determined fairly accurately. After irradiation, the yeast was permitted to grow for a short time in order to bring out the growth rate differences, and was then compared with the controls.

By this method, KANNEGIESSER (1931) working with yeast blocks, each representing approximately 50 Å, studied three types of oxidation, namely pyrogallic acid in alkaline solution by air, glucose + $KMnO_4$, and blood serum + H_2O_2. It was found that the growth was stimulated only on the two blocks receiving radiation from 2220—2280 and 2280—2340 Å. None of the other detector blocks differed appreciably from the controls.

From these results, it would appear that all three oxidations gave the same spectrum, as far as can be ascertained with this rather crude method.

The next advancement was the division of the spectrum into separate strips of exactly 50 Å each. **Potozky** (1932), by means of glass needles and heavy cellophane, prepared a chamber (fig. 23) the sections of which corresponded exactly to the 50 Å divisions of her spectrograph. By this simple instrument, **Braunstein** and **Potozky** (1932) showed that the spectra of different oxidations possessed certain specific regions besides the general oxidation spectrum. The data of 7 separate experiments are reproduced in Table 11. The various spectra are not quite identical, and even the rather crude determination by 50 Å strips shows differences which remained typically constant when the experiments were repeated. The limit of error is ± 15.

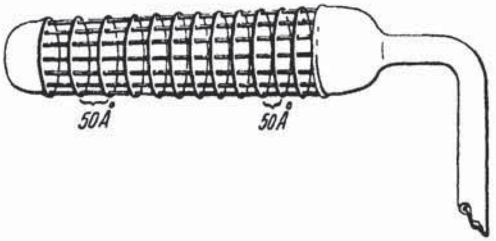

Figure 23.
Device for exposing yeast to successive ranges of the spectrum of 50 Å each.

Table 11. Induction Effects obtained from 50 Ångstrom strips of the Spectra of various Oxidations

	Detector: Cell Numbers in liquid Yeast Cultures										
	1900—1950	1950—2000	2000—2050	2050—2100	2100—2150	2150—2200	2200—2250	2250—2300	2300—2350	2350—2400	2400—2500
$KMnO_4+H_2O_2$	+10	+8	+1	+18	+30	+18	+63	+18	+70	+ 2	+5
$K_2Cr_2O_7+FeSO_4$ . . .	+11	+8	+4	+ 2	+30	+23	+68	0	+45	0	+1
$HNO_3+FeSO_4(+H_2SO_4)$	+ 5	—7	+3	+ 5	+ 4	+ 2	+10	+19	+29	+34	—7
$KClO_3+Zn+NaOH$ . .	+ 7	0	0	+ 4	+ 8	+20	+23	+26	+28	— 5	+2
$FeCl_3+NH_2OH \cdot HCl$.	+ 5	—2	+3	+ 7	— 9	0	+46	+78	+90	+51	—2
H_2O_2+Pt	+ 6	— 3	0	+13	+29	+39	+29	+20	+36	— 8	+1
$HgCl_2+SnCl_2$	+ 4	+6	—2	+ 3	+49	— 4	+60	+58	+97	+36	—4
								Range obtained by Kannegiesser			

It was also found that diffuse daylight increases the intensity of some reactions, e. g. of $K_2Cr_2O_7 + FeSO_4$, but does not affect the spectrum itself.

A still more detailed analysis was finally accomplished by PONOMAREWA (1931) who used 10 Å sections. It was impossible to divide the agar surface into such narrow strips and there was always the possibility of confusion by the spreading effect (see p. 110). Therefore, PONOMAREWA screened off all radiation except one narrow slit of 10 Å. The position of this slit could be changed over the entire spectral range By this method, only one small part could be studied at one time, and the progress of such analysis is slow. However, if the general spectrum has been investigated by the above-mentioned coarser methods, the negative regions need not be examined, and the amount of work is thus greatly reduced. On account of the very low intensity, this procedure is usually combined with intermittent radiation (see p. 103). In this way, PONOMAREWA could show that the glycolytic spectrum of blood consists of only 5 regions, or, more precisely, that only 5 of the 60 spaces of 10 Å each manifested mitogenetic effects (see fig. 24). There may be more than one spectral line, of course, in a strip of 10 Å. Recently, DECKER (1934) succeeded to to split the first double line of this spectrum into two different lines of 5 Å each.

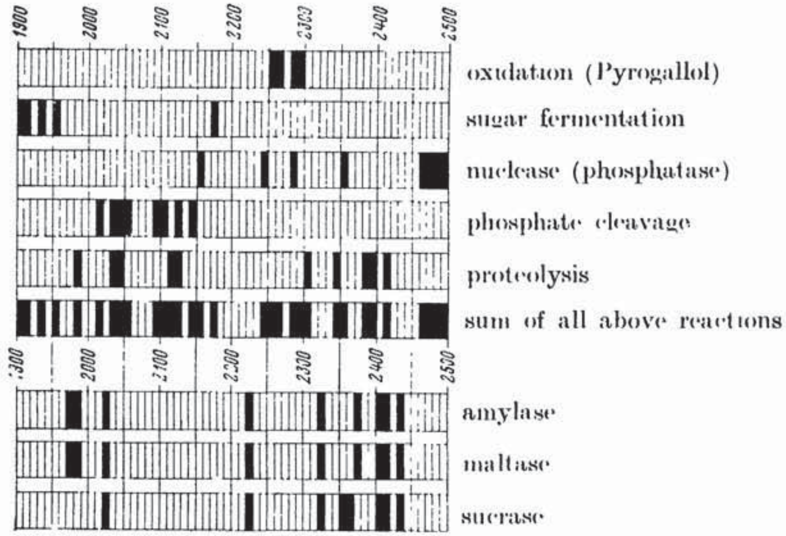

Figure 24. The spectra of some common biological reactions.

The most important result, however, was the observation that these bands coincided exactly with those of the alcoholic fermentation by yeast, and also with those of the lactic fermentation by Streptococci. GURWITSCH concludes that there must be some process common to all of these sugar decompositions, giving off the same radiation. This seems probable since it is generally assumed that the cleavage of the hexose phosphate through glyceric aldehyde to methyl glyoxal is common to all three types of sugar decomposition.

As a consequence of these splendid findings, the method was used for a number of frequently-occurring biological reactions. After some preliminary analysis by LYDIA GURWITSCH (1931), BILLIG, KANNEGIESSER and SOLOWJEW (1932) produced the detailed proteolytic spectrum. Two sets of data were obtained, one with the digestion of serum albumin by the gastric juice of a dog, and another from the splitting of glycyl-glycine by erepsin. The two spectra proved to be exactly alike. The authors assume that the source of radiation is the deaminisation of the amino-acid group (see also Table 23 p. 74).

The splitting of nucleic acid by the pulp of adeno-carcinoma of a mouse has a spectrum decidely different from that of proteolysis. It was determined by A. and L. GURWITSCH (1932a) and is also shown in fig. 24 together with that of glycolysis and of an oxidation. The "nuclease" gives a very long wave length. The same lines have been found by GURWITSCH in the decomposition of lecithin by "lecithase" (unpublished; quoted from BRAUNSTEIN and SEVERIN, 1932). Since both enzymes split the phosphoric acid radical from the organic remainder, the Russian school now calls this the "phosphatase spectrum".

BRAUNSTEIN and SEVERIN (1932) attempted to analyze the spectrum of a different type of organic phosphate cleavage, namely that of amino-groups coupled with phosphoric acid in phosphagen. This, according to LUNDSGAARD, plays an important role in the energy for the working muscle. They prepared Ca-creatin phosphate from muscle, and its chemical decomposition by means of H_2SO_4 was the source of radiation. The spectral analysis was carried out by counting the total number of yeast cells (method, see p. 72). The final determination, in 10 Å strips, showed 8 with definite radiation. The line 2000—2100 is doubtful, and was considered negative by these authors, but has subsequently been assumed as positive in all Russian publications. The mitogenetic spectrum of the working muscle (FRANK, 1929) contained some lines which at the time could not be accounted for, and which now can be explained by this phosphate cleavage.

These are the detailed spectra of simple chemical reactions published at present, as far as we have been able to ascertain. As oxidation spectra, the two double lines of pyrogallol oxidation have been inserted; these are commonly used by the Russian workers for this purpose. Fig. 24 shows that only rarely is the same line produced by two different processes. Even then, it should be realized that we are not dealing with true spectral lines, but with relatively broad regions, and that two identical strips do not necessarily indicate two identical lines, but rather two (or more) proximate lines.

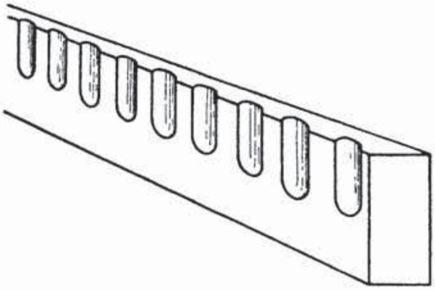

Figure 25.
Grooved glass block for exposing bacterial cultures to the various wave lengths of the spectrum, to be used with quartz plate in front, taking the place of the photographic plate in the spectrograph.

There is also a spectrum shown which is the sum of all these processes. We shall see later (p. **156**) that the spectra of nerves frequently combine all of these lines, in addition to some others of unknown origin.

The spectra of the action of amylase and maltase, and of sucrase (invertase) have been obtained by KLENITZKY and PROKOFIEWA (1934). The lines of these two enzymes agree to a much larger degree than any of the previously-mentioned processes. This is to be expected from their chemical parallelism.

Another method of obtaining spectra is that of WOLFF and RAS (1932) who used bacteria as detectors. They made a number of vertical grooves in a glass block which fitted into the camera of the spectrograph (see fig. 25); the grooves were covered by a quartz plate so that they became tiny pockets into which the detector culture was placed for exposure. The wave length for each one could be determined accurately.

By this procedure, they found the spectrum of neutralization of acid and alkali to consist of three lines

a strong line between 1960 and 1990 Å
a strong line between 2260 and 2300 Å
a weaker line between 2070 and 2090 Å.

Fig. 26 shows these regions, and also the spectra obtained by Ras (quoted from Ruyssen, 1933) for the Bunsenburner flame. This latter spectrum has also been photographed, and had many more lines of longer wave length, some of which are shown here, but photography failed entirely at the shorter ultraviolet. — The figure also shows the biologically obtained spectrum of the reaction of hydrogen with chlorine.

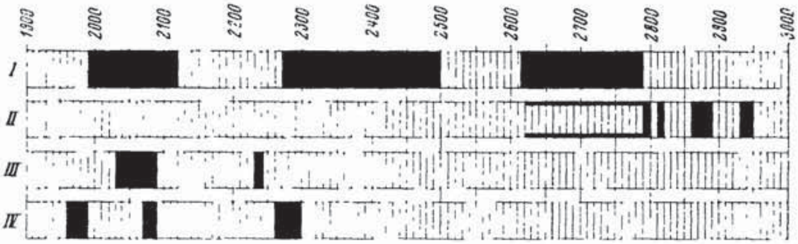

Figure 26. Photographic and mitogenetic spectra:
I. Bunsen burner flame, mitogenetic spectrum; *II.* same, photographic spectrum; *III.* Reaction $H_2 + Cl_2$, mitogenetic spectrum; *IV.* Reaction $NaOH + HCl$, mitogenetic spectrum.

It should not be gathered from this discussion that the indicated lines represent the entire spectrum of these reactions. On the contrary, it is most probable that it extends to both sides of the narrow range shown here. We are limited, however, by our indicators. Radiation below 1900 Å will be readily absorbed even by air (see p. 23), and that above 2600 Å does not produce mitogenetic effects (see fig. 28); therefore they can not be observed by the methods employed here. They may be capable of producing other biological effects hitherto unaccounted for, and may play an important part in chemical reactions.

A good summary of the Russian studies of mitogenetic spectra, including those of inorganic reactions, and with sufficient data to obtain a conception of the error of the method, has been given by A. and L. Gurwitsch (1934).

The intensity of ultraviolet radiation from a chemical reaction is not proportional to the total energy liberated. This was to be expected because the same holds true for the emission of visible light from chemical reactions. Strong mitogenetic effects can be obtained from protein digestion by pepsin or from milk coagulation by rennet while Rubner found the heat of reaction of proteolysis to be immeasurably small. On the other hand, the heat of neutralization of acid by alkali is so large that it can be observed even without a thermometer, yet its radiation is relatively weak.

The origin of the spectra is by no means clear. Lorenz's criticism (1934) can be explained in a concrete example as follows: If a spectrum represents the radiant energy emitted by the reaction as such, then each reacting molecule must emit at least one quantum of the shortest wave length observed in the spectrum. The longer wave lengths might originate from the shorter ones by loss of definite amounts of energy. Thus, in the case of sucrose hydrolysis by invertase (figure 24) each sucrose molecule must emit at least one quantum of the wave length 2020 Å, i. e. of the energy content 9.8×10^{-12} ergs (see Table 1). There are 6.1×10^{23} molecules in a grammolecule, or $\frac{6.1 \times 10^{23}}{342}$ in 1 g of sucrose. The minimal amount of radiant energy from 1 g of sucrose would then be:

$$\frac{6.1 \times 10^{23} \times 9.8 \times 10^{-12}}{342} \text{ ergs} = 0.175 \times 10^{11} \text{ ergs} = 0.042 \times 10^{4} \text{ cal} = 420 \text{ calories.}$$

The total energy liberated by this hydrolysis has been measured by Rubner (1913) by means of a Beckmann thermometer in silverlined Dewar bottles, and was found to be 9.7 calories per gram. This experimental value is only one-fiftieth of the minimum amount calculated. It seems impossible that appreciable amounts of energy could have been lost by the method used. We are compelled to the following alternative: Either, the spectra do not originate from the reactions for which they are considered specific, but from some quantitatively unimportant side reaction; or, the calculated amounts are actually liberated, but are at once absorbed again.

This latter explanation does not appear entirely impossible. The biologist is familiar with "false equilibria", such as the stability of sugar in the presence of air, though it could be oxidized with liberation of much energy, and the process might, therefore, be expected to take place spontaneously. Modern chemistry explains this by the necessity of "activation" of the molecule to make it react chemically. This activation requires energy. **Fricke** (1934) in an introductory summary, makes the following general estimates:

"Generally, energies of activation are of the order of 100,000 gram calories per gram molecule which equals 1.5×10^{-19} gram calories per molecule. At ordinary temperature, the average kinetic energy of a molecule is of the order of 10^{-21} gram calories. Only the inconceivably small fraction of 10^{-13} of the molecules have energies in excess of 10^{-19} gram calories per molecule.

"The quantum theory gives as the reason that activation may be produced by radiation, the fact that the energy of the radiation is carried in a concentrated form, as quanta. The energy of a quantum of radiation is $5 \times 10^{-16}/\lambda$ gram calories where λ is the wave length in the **Ångstrom** unit. The wave length has to be reduced to the order of 3,000 Å. before the quantum has the value 1.5×10^{-19} gram calories which, as we saw, represents the usual value for the energy of activation per molecule."

The energy of activation necessary for the hydrolysis of sucrose has not been determined. If we assume it to be of the "usual value" as computed by **Fricke** it would be equivalent to a quantum of about 3,000 Å. wave length per molecule. When this is absorbed, the sucrose molecule hydrolyses, and the amount of energy thus released must be larger than that absorbed, because of the additional heat of reaction of 9.7 calories per gram. The free energy will be absorbed at once by a neighboring sucrose molecule which becomes activated, and hydrolyzed, and thus the reaction goes on. In this way, all the liberated energy is again absorbed, except for the difference between heat of hydrolysis and heat of activation which we measure as heat of reaction.

However, there is another small "leak". Of those molecules adjacent to the walls of the vessel, the energy may radiate into the wall rather than to another sucrose molecule, and these few quanta would leave the system, and produce a radiation if the vessel is transparent. The amount of energy thus leaving the vessel would be extremely small, and would be of a wave length equal or shorter than that required for activation.

If this explanation of the mitogenetic spectra is correct, they would be an excellent means to measure the energy of activation.

C. SECONDARY RADIATION

A phenomenon must be recorded here which was first believed to be typical of living organisms, but is a property of certain chemical solutions, or systems, namely the emission of rays from a solution as a response to "primary" rays directed upon it.

The first purely chemical effect of this kind was observed by A. and L. GURWITSCH (1932b) with nucleic acid. A 3% solution of nucleic acid was gelatinized in a glass trough. At one end, it was irradiated with the lines 3220--3240 Å from a copper arc, through a monochromator. At the other end of the trough, radiation of the nucleic acid could be observed, but the wave length of this "secondary" radiation was not the same as that of the primary: it was between 2450 and 2500 Å, which is the spectrum of nucleic acid hydrolysis (see fig. 24). This proves that it can not be merely a reflection of light, because the wave length changed, nor is it a case of fluorescence, for the secondary radiation has a shorter wave length than the primary. And most remarkable of all, the induced light may be stronger in intensity than the primary source.

The best explanation is most probably the one given by GURWITSCH that the primary radiation induces some kind of chain reaction (see p. 47) which is then radiating with its own spectrum. This would account for the difference in wave lengths as well as for the increase in intensity.

Other examples have been given by WOLFF and RAS (1933b). These authors observed the same effect with sterile blood serum. They found also that sterile nutrient broth did not produce secondary radiation, but showed it when bacteria had grown in it, even after the bacteria themselves had been removed by filtration through a porcelain filter. A very short action of living bacteria suffices to change the broth to a "secondary sender". These filtrates as such emit no primary radiation. WOLFF and RAS observed further that after long exposure to primary radiation, these liquids ceased to produce secondary radiation. Very intense primary light caused a more rapid exhaustion. 45 minutes exposure of a staphylococcus supsension to a strong primary sender had made it unfit to produce secondary rays; however, on the next day, the suspension reacted normally again. In another case, even 30 minutes exposure was sufficient to destroy the power of secondary radiation.

Nucleic acid solutions also cease to function when overexposed, and during this stage, they do not transmit mitogenetic rays. Strong solutions recover again after 1 or 2 days of "rest".

An important observation is that with increasing concentration of the acting substances, the secondary radiation becomes weaker. Table 12 represents an experiment with nucleic acid, by Wolff and Ras. The source of primary radiation was the reaction of milk with rennet. The intensity of secondary radiation from the nucleic acid dilutions was measured by the length of exposure required to produce a "mitogenetic effect", i. e. to accelerate the growth of bacteria. The numbers in the table represent the percentage increase in cells over the control.

Table 12. Intensity of Secondary Radiation of Nucleic Acid solutions, measured by the time required to produce a "mitogenetic effect"

Concentration of Nucleic Acid, in %	Mitogenetic Effect after Irradiation for										
	15″	30″	1′	1.5′	2′	3′	4′	5′	6′	7′	8′
1.000							0	32	0	0	0
0.750							40	26	0	0	
0.500					33	55	12				
0.250		0	27	21	0		0				
0.100		0	43	17							
0.020		25	32	0							
0.004	0	0	0								
0.001	0	0	0								

The results are somewhat surprising. The lowest efficient concentration was 0.02%, which produced a mitogenetic effect at least 5 times as strong as the 1% solution, i. e. it produced the same effect in one-fifth the time. A similar relation was observed with bacterial suspensions. The more dilute they are, the stronger is the secondary radiation they emit upon excitation by some primary source. Perhaps this is brought about by the absorption of rays in overexposed solutions (see above).

Bacterial suspensions and their filtrates lose the power of secondary radiation upon heating; nucleic acid solutions do not.

The increase in intensity by secondary radiation enabled Wolff and Ras to construct an "amplifier" for mitogenetic rays.

They placed six quartz cuvettes filled with staphylococcus suspension side by side, and by irradiating one side of the series, obtained a good mitogenetic effect from the opposite side in 10 seconds; the same primary source used for direct irradiation of the same detector required 4.5 minutes. This means an amplification of 27 times. The observation is added that one continuous column of the same length as all 6 cuvettes together, does not give a greatly increased intensity.

It must be kept in mind that the intensity of radiation has been ascertained only biologically, by the time required to produce a mitogenetic effect. It has already been pointed out (p. 24) that the reciprocity law (requiring double exposure time for half the intensity) does not even hold for such simple reactions as those in the photographic plate when the intensity becomes very low. Its application to biological reactions is quite doubtful. Recently, WOLFF and RAS (1934a) could show that usually, secondary radiation is polarized, and they also proved that polarized mitogenetic rays exert a very much stronger effect upon organisms. It is not at all certain, therefore, that the above statements really indicate an increase in intensity of radiation in the physical sense of the word.

In his most recent summary, GURWITSCH (1934) makes the following statement:

> "It has been found that all substrates capable of enzymatic cleavage, such as glucose, proteins, nucleic acid, urea, fats etc. react also upon mitogenetic radiation and become radiant. This "secondary radiation" has certain properties of great interest. (1). it travels from the irradiated part through the liquid medium to distances of several centimeters with the measurable speed of a few meters per second; and (2). the radiation is resonant, i. e. the substrate reacts mostly upon those wave lengths which it emits when decomposed enzymatically."

The only published experimental proof for this is that by DE KORÓSI (1934) as far as the authors have been able to ascertain.

CHAPTER III

EFFECT OF ULTRAVIOLET RADIATIONS UPON CELLS

A. EFFECT OF RADIATIONS UPON CHEMICAL REACTIONS

It has long been known that energy in the form of visible light, or near this range, will produce chemical changes, which we term photochemical reactions. Just as a chemical synthesis may be brought about by heat, so may it be induced by adding radiant energy in the form of light. All organic matter is thus produced from CO_2, H_2O and nitrates, by means of radiant energy from the sun, with chlorophyl as the necessary "transformer" of the energy.

It is not necessary, however, that radiant energy be present to increase the energy level of each reacting molecule. A number of chemical reactions are known which require some radiant energy to be initiated; however, several hundred, or even several million molecules are changed for each energy quantum which is absorbed. These reactions are exothermic. If only one molecule becomes activated by an energy quantum of the right size, the energy liberated by its reaction activates another molecule. Thus, many molecules may be changed though it is quite impossible that one quantum can be absorbed by more than one molecule. This type of reaction, which is called a *chain reaction*, is usually explained in the following way (H. S. TAYLOR, 1931, p. 1009—1018):

The quantum, in being absorbed by a molecule, ionizes it. The ion pair thus produced initiate two independent reactions which again produce ions. The classical example by BODENSTEIN and NERNST is the photochemical reaction between the gases H_2 and Cl_2. The chain reaction can be written

$$\mathrm{Cl_2 + h\nu = Cl + Cl}$$

$$\mathrm{Cl + H_2 = HCl + H} \qquad\qquad \mathrm{Cl + H_2 = HCl + H}$$

$$\mathrm{H + Cl_2 = HCl + Cl} \qquad\qquad \mathrm{H + Cl_2 = HCl + Cl}$$

$$\mathrm{Cl + H_2 = HCl + H} \qquad\qquad \mathrm{Cl + H_2 = HCl + H}$$

$$\mathrm{H + Cl_2 = HCl + Cl} \qquad\qquad \mathrm{H + Cl_2 = HCl + Cl}$$

$$\mathrm{Cl + H_2 = HCl + H}$$

$$\mathrm{H + Cl = HCl.}$$

Under "chain" is understood the number of consecutive molecules entering into reaction. The chain may be as "long" as one million molecules The "length" can be ascertained by determining the number of molecules changed for each quantum of light entering the system. The chain is terminated by the reactive molecules or atoms combining with each other, as indicated in the above model, or by reacting with other molecules to form stable compounds.

If it were not for these terminations, one quantum would be sufficient to cause all molecules to react with one another. In fact, in the case of explosions, where the reaction liberates a large amount of energy, this is practically the result.

The presence of foreign substances reacting with the components of the system is a common cause of cessation. If there were only one such molecule present for every million molecules of hydrogen and chlorine, that would account for an average chain length of one million molecules. If there were 100 times as much of the foreign substance, the chain length would be reduced to ten thousand molecules.

The same reasoning holds true for chain reactions in solutions. In the photochemical oxidation of Na_2SO_3 to Na_2SO_4, the quantum yield was about 100000. This reaction was inhibited by primary and secondary, but not by tertiary alcohols. Whenever a chain was terminated, two molecules of the alcohols were oxidized to the corresponding aldehydes and ketones.

The known chain reactions are exothermic. It does not seem imperative, however, that they be so if another source of energy is available. This is the case in normally nourished cells of animals, fungi, bacteria, etc., which liberate energy constantly by metabolizing carbohydrates, fats, proteins or other organic compounds. By means of this energy, they grow, i. e. they synthetize new body substance endothermically.

It is not impossible, that very small amounts of energy, even a single quantum, might produce a very noticeable effect in a living cell. This could be accomplished by releasing a complex mechanism which needs only one or several quanta of a given size to be initiated, just as the Cl_2-molecule needed only one quantum of the right size to start the reaction with hydrogen. The most common, and perhaps the only reaction in the cell which can be thus released is cell division; this results in a more rapid multiplication, or an increased growth rate.

B. EFFECT OF MONOCHROMATIC ULTRAVIOLET UPON LIVING CELLS

Ultraviolet light of any of the different physical sources mentioned in the previous chapters may have a very distinct effect upon living cells. The best-known is the reddening of the skin by ultraviolet light. A quantitative study of the relation between the intensity of the effect and the wave length has revealed that aside from the very marked erythema produced by wavelengths around 3000 Å, the cells of the skin will also react upon those below 2600 Å which are not found in sunlight. Between these two maxima is a zone of very weak effects. This fact is significant because the major part of this book is concerned with mitogenetic rays which are shorter than 2600 Å.

Ultraviolet light also kills bacteria and other microorganisms. Strangely, however, the intensity curve for the different wavelengths is quite different from that of the erythema effect, it looks almost like the reverse. Figure 27 shows the results by **Coblentz**, **Stair** and **Hogue** (1932) on erythema, by **Rivers** and **Gates** (1928) on vaccine virus and *Staphylococcus aureus* and by **Duggar** and **Hollaender** (1934) on *Bact. prodigiosum* and the mosaic virus of tobacco. Most of these investigations have been carried out no further than to a wave length of about 2500 Å.

If shorter wave lengths are taken into consideration, and especially if the intensity is very greatly decreased, it is possible to obtain growth stimulation under certain conditions which will be specified in Chapter IV. The result of the most extensive of the many experiments of this nature is shown graphically in fig. 28.

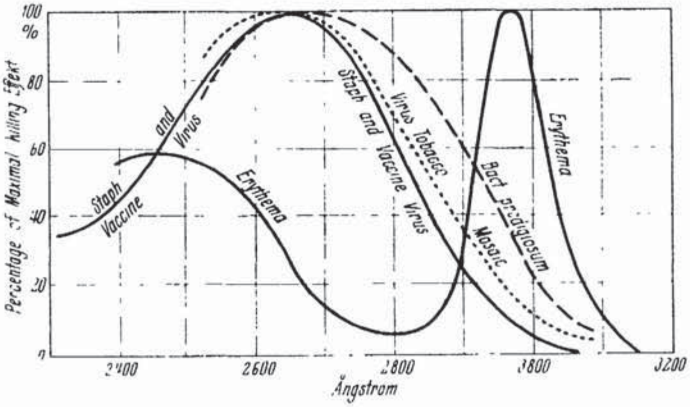

Figure 27. Comparative intensities of the killing effect of different wave lengths. The intensities are uniform for each individual organism, but vary greatly for the different curves.

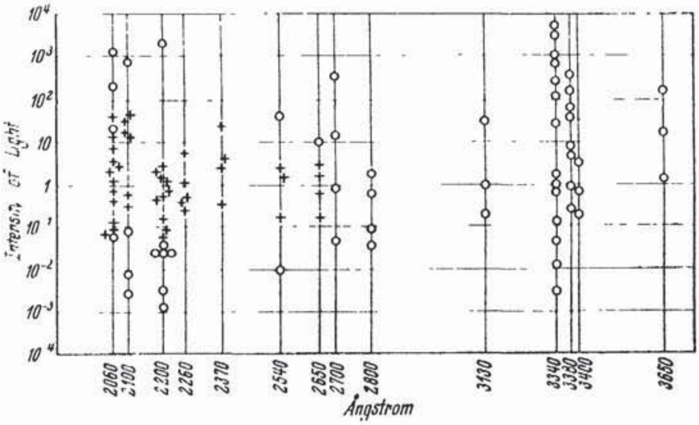

Figure 28. The effect of ultraviolet light of different wave lengths and different intensities upon yeast.

+ indicates accelerated growth; o means no effect.

Chariton, Frank and Kannegiesser (1930) used individual spectral lines, employing a monochromator, from the sparks of aluminum, zinc or cadmium, to irradiate yeast cultures. Each culture thus obtained light of one definite wave length. By varying the intensity as well as the wave length, it could be shown that only radiation of less than 2700 Å produced positive effects, i. e. increased the growth rate of yeast.

A large number of similar experiments have been carried out by Gurwitsch and his associates, usually as controls for organic radiations. They will be mentioned in the succeeding chapters.

There seems to be very little difference in the limiting intensities of different wave lengths. Other experiments have shown that even at 1900 Å, good effects can be obtained. Below 1900 Å, absorption by quartz, water and air interferes with the experiment.

C. EFFECT OF RADIATION FROM CHEMICAL REACTIONS UPON LIVING CELLS

The same effect which has been demonstrated above as the result of irradiation with ultraviolet of known wave lengths, can be produced also by exposing the cells of microorganisms to the emanation from chemical reactions.

One of the simplest examples is the stimulation of the bacterial growth rate by the emanations from the neutralization of NaOH with HCl. Wolff and Ras (1933b) allowed these two chemicals, flowing from two tubes, to unite on a quartz plate, underneath which was the bacterial culture. After exposure, these cultures were incubated for 2 hours. Table 13 shows very distinctly in both experiments that an exposure of approximately 5 minutes to the radiation of the neutralization process has stimulated the growth; the number of cells has been increased approximately 40--50 %.

The same authors found that even the dissolution of NaCl in water produces a growth-stimulating radiation (Table 14). This energy emission occurs only during the act of dissolving; it ceases completely when all the salt is in solution. No effect is noticeable when sugar is dissolved in water, or when palmitic acid is dissolved in alcohol. Wolff and Ras concluded therefore that the process of dissociation of salt into ions is the source of ultraviolet.

Table 13. Staphylococci exposed through quartz to the energy emanations of the reaction $NaOH + HCl = NaCl + H_2O$ and counted 2 hours later

Exposed for	Cells per cc. of culture	
	I	II
0 minutes (control)	26 100	26 800
4 ,, ...	26 200	31 500
4.5 ,, ...	27 250	
5 ,, ...	37 750	39 500
6 ,, ...		30 900
each number is the average of	9—11 experiments	3—4 experiments

In the same way, bacterial growth was accelerated by exposure to metallic zinc in a solution of lead acetate or copper sulphate.

Simple oxidation processes also emit energy which can cause an increase in the growth rate of bacteria or yeasts. This is already cited in the method of obtaining oxidation spectra, and may be further illustrated by an unpublished experiment of Miss A. J. Ferguson. Oxalic acid was oxidized with permanganate in a glass vessel. Above this were fastened two small covered dishes, one of quartz and one of glass, each containing a sample of the same culture of *Bacterium coli*. The sample in the quartz vessel grew more rapidly, having been exposed to ultraviolet light from the oxidation process; the other received no stimulus since glass

Table 14. Staphylococci exposed through quartz to the energy emanation from dissolving substances

Duration of Exposure	Cells per cc. of culture								
	0 Control	10″	20″	30″	45″	1′	1.5′	2′	4′
NaCl in water	32 100	31 400	37 700	45 000	—	—	—	—	-
	21 750	—	—	31 500	27 300	-	—	—	-
	35 500	- -	47 500	45 500	-	40 000	36 800	—	-
sucrose in water	7 000		-	6 250	—	7 250	7 250	7 250	-
palmitic acid in alcohol	7 000	—	—	7 000	-	8 000	8 500	7 250	7 500 7 750
NaCl after complete solution	21 750	—	—	22 000	20 750	—	—	—	—

absorbs the radiation (Table 15). There is the usual lag period of 2 hours, but after the bacteria once start to grow, the irradiated culture grows more rapidly.

Table 15. Development of a culture of Bacterium coli after exposure to emanations from the reaction
$C_2O_4H_2 + KMnO_4 = K_2CO_3 + 2\,MnO + 9\,CO_2 + 5\,H_2O$

	Cells per cc. of culture	
	exposed through glass	exposed through quartz
Immediately after exposure .	149	149
1 hour later	154	140
2 ,, ,, ...	253	216
3 ,, ,, ...	940	1735
4 ,, ,, ...	3335	9085

What holds true for the simpler chemical reactions, is also correct for the more complicated biochemical processes. Proteolysis by enzymes yields an ultraviolet emanation which greatly stimulates the growth of yeast as seen in Table 16, containing the data obtained by Karpass and Lanschina (1929). Of 12 experiments, only one was negative.

Table 16. Increase in the development of yeast cultures after exposure to the emanation from proteolytic processes

Proteolytic process	Percentual increase of exposed culture over control
Egg white with pepsin.	43 %; 20.9 %
egg white with pancreatin	10.7 %; 25.8 %; 15.5 %
egg yolk with pepsin	20.6 %
egg yolk with pancreatin	30.1 %; 31.4 %
fibrin with gastric juice	37.1 %; 36.6 %; 20.4 %; −16.5 %

All other enzymic processes which have been tested so far have yielded positive growth stimulation. Since all organisms display processes liberating energy, it is only logical to assume that all living organisms radiate. This statement must be modified somewhat by the consideration that these ultraviolet rays are very readily absorbed, and, for example, will not pass the skin of man or animals. Which parts of the various animals and plants radiate, will be discussed in Chapters IV and VII. Attention should be called here only to the fact that there may be radiations and growth stimulation inside of an organ or tissue without becoming noticeable outside this focus. Since we must expect ultraviolet radiations from very many biochemical processes, and since they may stimulate cell division they may play an extremely important role in the development of all living beings.

CHAPTER IV

METHODS OF OBSERVING BIOLOGICAL RADIATIONS

The preceding chapters have shown that for physico-chemical reasons, we should expect ultraviolet radiations from all living organisms, as long as they have any noticeable metabolism. In this chapter, the methods used in detecting and proving such radiations will be discussed. By far the most extensive treatment is given to mitogenetic radiation because it is the most studied and the best understood. The necrobiotic rays and the injurious human radiations are, perhaps, only special manifestations of mitogenetic radiation. The Beta-radiation of living as well as dead organisms is only mentioned in passing.

The presentation in this chapter is largely historical. Occasional exceptions to this arrangement could not be avoided.

A. MITOGENETIC RADIATION

This type of ultraviolet rays was discovered by Gurwitsch in 1923. He called them mitogenetic because he observed that they stimulated cell division, or mitosis. This radiation is so weak that it was not possible for a long time to verify its existence by physical measurements. Its effect upon living organisms is very conspicuous, however. In onion roots, it increases the number of mitoses. It accelerates the growth of yeasts and bacteria, the development of eggs, and the division of certain cells in the animal body. It may cause morphological changes in yeasts and bacteria, and in the larvae of sea urchins.

a) The onion root method

The root of an onion, used by Gurwitsch and his associates extensively from 1923 to 1928, was the first detector of the rays. The detector root was placed in a narrow glass tube to permit easy handling. The zone of meristematic growth, a few mm from the tip, was uncovered allowing it to be irradiated. In the first experiments (1923), the source of radiation was another onion root, also placed in a glass tube so that it could be directed to point exactly at the growing tissue of the first root (fig. 29). The roots were left in this position for one or two hours. Two to three hours after the beginning of irradiation, the detector root was fixed and stained, and, in microtome sections, the number of dividing nuclei was ascertained. GURWITSCH found that the side of the root exposed to the biological radiation showed regularly more dividing nuclei than the opposite side. For this reason, GURWITSCH coined the word "mitogenetic" rays. Table 17 gives the results of a test with the crushed base of an onion (A. and L. GURWITSCH, 1925).

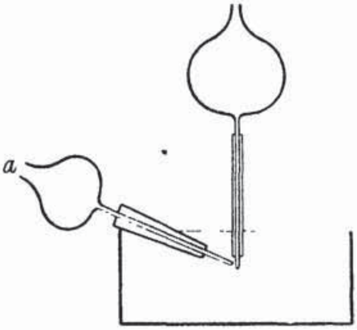

Figure 29.
The arrangement of the onion roots in the first experiments on mitogenetic rays.

Table 17. The radiation of onion base pulp

The numbers given are those of dividing nuclei in corresponding microtome sections of the exposed and the unexposed side of the detector root.

	A: fresh pulp													Totals
exposed	51	59	90	82	61	60	57	53	72	62	59	37	45	788
unexposed	49	63	60	50	51	38	45	43	57	47	38	30	30	601
difference	2	−4	30	32	10	22	12	10	15	15	21	7	15	187 = +23.8%

	B: same pulp heated to 60° C.																Totals
exposed	60	66	64	61	61	63	60	75	81	71	61	71	73	73	65	61	1066
unexposed	61	69	65	62	74	62	60	73	82	66	65	74	69	69	65	60	1076
difference	−1	−3	−1	−1	−13	1	0	2	−1	5	−4	−3	4	4	0	1	−10 ~~= −0.9~~%

This technique was used by Gurwitsch and a number of associates to search for mitogenetic rays in the entire organic world. It would scarcely be worthwhile to compile a complete list of organisms found to radiate. The following list contains the more important earlier observations, compiled by Gurwitsch (1929).

Radiating organisms and tissues

Bacteria *Bacterium tumefaciens*, Staphylococci (Magrou)
Bacterium murimors (Acz) (Sewertzowa)
Bacillus anthracoides, *Sarcina flava* (Baron)
Streptococcus lactis (Magrou)
Yeast (Baron, Siebert, Magrou)
Eggs of annelids
Eggs of sea urchins before the 1st division (Frank and Salkind)
before the 2nd and 3rd divisions (Salkind)
Egg yolk of chicken, only during the first two days of incubation (Sorin). After establishment of circulation system, radiation ceases
Embryos of amphibia in the morula stage (Anikin)
Plant seedlings: root tips, cotyledons, young plumulae of *Helianthus* (Frank and Salkind)
Potato tubers: leptom fascicles only (Kisliak-Statkewitsch)
Onion roots connected with the bulb (Gurwitsch)
Onion base pulp (A. and L. Gurwitsch, Reiter and Gabor)
Turnip pulp, 24 hours old (Anna Gurwitsch)
Young tadpole heads, pulp of tadpole heads (Anikin, Reiter and Gabor)
Blood of frog and rat (Gurwitsch, Sorin)
Blood of man (Siebert, Gesenius, Potozky and Zoglina)
Contracting muscle (Siebert, Frank)
Pulp from resting muscle + lactic acid + oxygen (Siebert)
Corneal epithelium of starving rats, but not of normal rats (L. Gurwitsch)
Neoplasms: carcinoma, sarcoma (Gurwitsch, Siebert, Reiter and Gabor)
Spleen of young frogs (Gurwitsch)
Bone marrow (Siebert)
Bone marrow and lymph glands of young rats (Sussmanowitsch)
Resorbed tissue: tails, gills, intestine of amphibian larvae during metamorphosis (Blacher, Bromley)

Regenerating tissue of salamander and angleworm (BLACHER, SAMARAJEFF)
Hydra: hypostom and budding zone, not other parts.

Non-radiating organisms and tissues

Tissues of adult animals except brain, blood and acting muscle (most tissues have later been found to radiate slightly)
Tadpoles over 2 cm. long
Chicken embryo after 2 days (blood radiates)
Blood serum (becomes radiant with oxyhemoglobin) (SORIN) (or with traces of H_2O_2) (ANIKIN, POTOZKY and ZOGLINA)
Blood of asphyxiated frogs
Blood of cancer patients
Blood of starving rats (becomes radiant with glucose) (ANIKIN, POTOZKY and ZOGLINA)
Active tissues with chloral hydrate
Active tissues with KCN

Further details will be given in Chapter VII.

The greatest early support to the establishment of mitogenetic rays was given through the extensive and thorough work of REITER and GABOR (1928). These two authors tested the onion root method, and verified especially the physical nature of the phenomenon; the radiant nature of this effect was thus fortified beyond doubt (see p. 59).

Considering the great claims which Gurwitsch and his associates made for their discovery, it was surprising that comparatively few biologists were sufficiently interested to repeat the experiments. Among these, some obtained negative results so consistently that they denied the existence of mitogenetic rays altogether, and considered the results of the Russian workers and of REITER and GABOR to be experimental errors. The most frequently quoted of these are SCHWARZ (1928), ROSSMANN (1928), and much later, in this country, TAYLOR and HARVEY (1932). Positive results were obtained by MAGROU (1927), WAGNER (1927), LOOS (1930), BORODIN (1930), and recently by PAUL (1933), and many Russian workers, in several different laboratories.

In order to decide whether the positive results could be considered experimental errors, SCHWEMMLE (1929) undertook a statistical investigation. He concentrated graphically all results published by GURWITSCH and the Russian school (about 200) by plotting the percentage increase or decrease of mitoses of the exposed over the unexposed side of the root, against the total number of mitoses counted. He distinguished only between "induced" and "not induced" roots. From the "not induced" roots, he could compute the probable error of the method. The error was ± 10% when 500 mitoses were counted, and decreased, of course, as the total number increased (fig. 30). All results which had been claimed to prove mitogenetic radiation were found outside the limits of error.

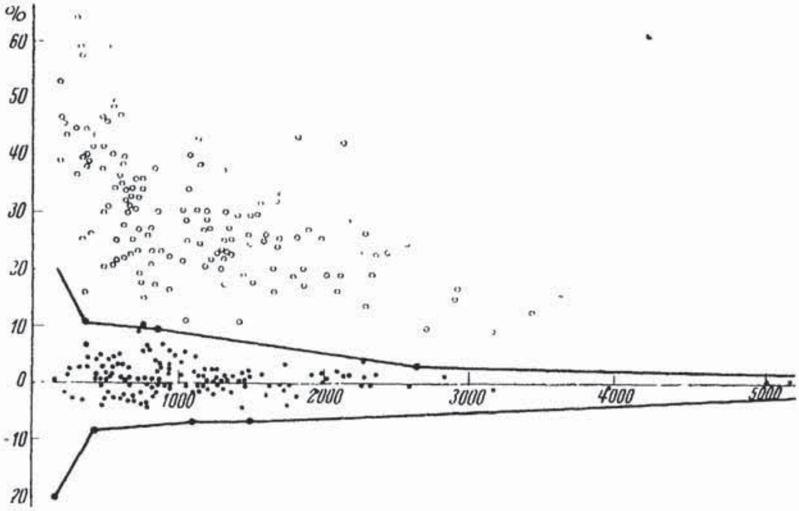

Figure 30. All results with onion root as detector by GURWITSCH and associates. Ordinate: percentage of increase over control; Abscissa: total number of mitoses counted. Circles indicate a positive induction, black dots indicate no mitogenetic effect. The line gives the limits of error.

REITER and GABOR's experiments have a much greater error, ± 20%, and a few experiments supposed by these authors to prove induced mitogenetic effect are really within the limits of error. The error of the experiments by WAGNER (1927), SCHWARZ (1928), and ROSSMANN (1928—29) is even larger than that of REITER and GABOR's, and TAYLOR and HARVEY's few experiments (1931) indicate a similar large error. All these data gave doubtful or negative results; the mitoses of two different sides of the roots varied greatly.

SCHWEMMLE did not consider it proved that the effect is caused by mitogenetic rays, because of the possibility of other physiological factors affecting mitosis during the experiments.

The effect as such, however, must be considered established by a very large number of data, and the only question was its interpretation. That under different physiological conditions, in different countries with different onions, negative results have been obtained by some investigators is not really surprising

Gurwitsch had always claimed that the effect of one root upon the other was caused by rays. In fact, he predicted in 1922 radiation as a factor in mitosis, and in trying to verify his prediction, found onion roots as the first reliable indicators.

The mitogenetic effect proceeds in a straight line and is reflected from glass and from a mercury surface (**Gurwitsch**, **Siebert**, **Reiter** and **Gabor**). It will pass through thin layers of quartz and of water (**Gurwitsch**, **Magrou**), through thin animal or vegetable membranes, thin plates of mica (**Reiter** and **Gabor**), thin cellophane (**Stempell**), but not through thick layers of glass, such as glass slides, nor through gelatin even in very thin layers. It seems hardly possible to account for all of this by any agent other than ultra-violet rays.

A very vital question is that of the wave length of these rays, and it is very interesting that two distinctly different wave lengths have been claimed, both based upon apparently reliable data.

Gurwitsch could obtain the effect through quartz, and partly through very thin glass, but not through very thin gelatin, and concluded that he was dealing with an ultra-violet radiation of about 2200 Å. **Frank** and **Gurwitsch** exposed onion roots to different wave lengths from physical sources, and obtained mitogenetic effects only from the spectrum between 1990 and 2370 Å.

Quite different were the results of **Reiter** and **Gabor** (1928). They found this radiation to be transmitted through 3 mm. of Jena glass, and still noticeably through 5 mm. of common glass, and also through gelatin which indicates a wave length above 3000 Å. By means of special filters, they found the range to be between 3200 and 3500 Å. Then, by irradiating roots with known wave lengths of the spectrum, they determined the mitogenetic efficiency of this part of the spectrum. Besides a sharp maximum at 3400 Å, another smaller maximum was discovered near 2800 Å. Below this, no mitogenetic effect was observed, not even in the neighborhood of 2000 Å which was considered by **Gurwitsch** and **Frank** as the only efficient region. The curve resembles somewhat that for erythema (fig. 27 p. 49).

The very interesting observation was made that the apparently inert spectrum between the two maxima will prevent mitogenetic effects by the active wave lengths, even when the intensity of the "antagonistic" rays is only one-tenth of that of the mitogenetic rays. All wave lengths between 2900 and 3200 A show this inhibition. The rays outside of the maxima were entirely neutral. Direct sunlight and ultra-violet arc light also inhibited mitogenetic effects.

Reiter and **Gabor** determined further the wave length of mitogenetic rays by means of a spectrograph, letting the spectrum from roots or sarcoma tissue fall upon the length of an onion root. All three experiments gave an increase in mitoses at the place where the wave lengths between 3200 and 3500 Å had fallen on the root.

These last experiments can now be explained by an error in technique. It was not known at that time that irradiation of the older parts of a root will produce a mitogenetic effect not at the place of irradiation, but at the only reactive part, namely the meristem near the root tip. This last argument in favor of a wavelength near 3400 Å must therefore be discarded. It is considered definitely established now that mitogenetic rays range between 1800 and 2600 Å, as has already been shown in Chapters II and III. However, the deviating experiences of **Reiter** and **Gabor** have never been accounted for in a really satisfactory way (see **Gurwitsch**, 1929).

The publication of **Reiter** and **Gabor**'s experiments caused the Russian workers to repeat them at once, because they contradicted all their own statements about the wavelength, and none of the German authors' results could be verified.

The first experiment was **Frank**'s spectral analysis (1929) of the radiation of the tetanized muscle, with a spectrograph using yeast as detector (see p. 35). In three well-agreeing experiments (fig. 31), it could be shown that no radiation above 2400 Å was emitted. Then followed the detailed study by **Chariton**, **Frank** and **Kannegiesser** (1930) of the effect of monochromatic light from physical sources upon yeast. The results have already been shown in fig. 28. Beyond 2600 Å, no variation of intensity produced any effect. Special efforts were made to investigate the range around 3400 Å claimed to be so efficient by Reiter and Gabor, but it yielded only consistently negative results.

Considering that Reiter and Gabor used onion roots as detectors, the experiments were repeated with onion roots. Again, the shorter wave lengths were found to be efficient, and those in the region of 3400 Å gave no effect (see Table 18).

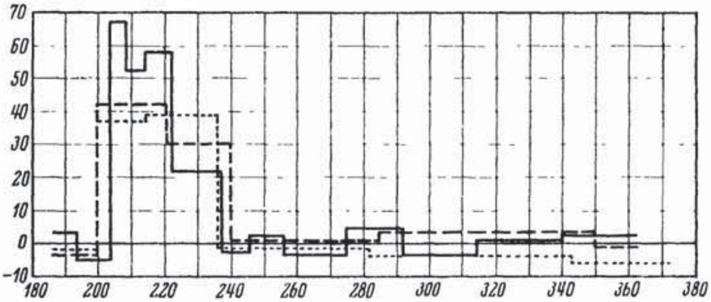

Figure 31. Results of 3 experiments on the spectrum of muscle radiation, with the technique shown in figure 22. Since the width of the agar blocks was not uniform, the results overlap partly, but show the general spectrum.

By these and other methods with a variety of indicators, the wave lengths of various radiations have been shown to be quite different, but all of them were below 2600 Å No records are

Table 18. Irradiation of Onion Roots with Monochromatic Spectral Light

Efficient Wave Lengths according to					
Gurwitsch			Reiter and Gabor		
Wave Length Å	Relative Intensity	Induction	Wave Length Å	Relative Intensity	Induction
2190	0.02	+24	3340	0.20	-1
2350	0.02	+20	3340	0.40	+5
2350	0.10	+30	3340	2.00	2
2350	1.00	+25	3340	80.00	0
			3340	800.00	—1
			3380	0.03	—7
			3380	4.00	+8
			3380	60.00	—2

given of wave lengths below 1900 Å. This is no proof that they do not exist, but rather, that below this point, absorption by quartz and air in the spectrograph makes accurate determinations impossible. It can hardly be doubted from the large amount of spectra analyzed by the Russian school, and confirmed by WOLFF and RAS (p. 40), that mitogenetic radiation consists essentially of the wave lengths between 1900 and 2500 Å.

Since 1928, the onion root as detector has been substituted by the time-saving yeast methods or by bacterial detectors. However, two extensive recent investigations must be mentioned which concern the question whether onion roots can be used at all as detectors. Both papers describe the technique employed very carefully, and they arrive at quite different conclusions.

MOISSEJEWA (1931, 1932) observed that roots when removed from the water show symmetrical distribution of mitoses, but after repeated removal, they do not. Pressing or rubbing of the roots will increase the number of mitoses, and after long continued pressing the opposite effect occurs. When friction and pressure were carefully avoided, no increase of mitoses was observed upon exposure to another onion root.

MOISSEJEWA denies the existence of mitogenetic effects in onion roots and explains GURWITSCH's consistent results by several assumptions: (1) One-sided pressure of curved roots in the glass tube. (2) Light applied repeatedly in centralization of roots which causes phototropic curving of the root and results in increased mitosis. (3) Selection of good roots for important experiments which results in increased mitosis through pressure, and of less uniform roots when no effect is expected, which then leads to negative results. (4) Omission by GURWITSCH of the microtome sections showing a decreased number of mitoses when they happen to come between sections showing an increased number. — With yeast and blood as senders, this author obtained also negative results.

This careful study has been considered by many critics to be the final proof against mitogenetic radiation. Most of them do not mention the still more careful work by MARGARETE PAUL (1933). Realizing the prompt reaction of roots to touching, PAUL fastened the small onions (hazelnut size) by means of gauze to perforated cork stoppers; these were held above the ground by simple stands; in a completely covered moist chamber, in a dark room at 20° C, the roots developed in the air through the muslin in two to five days, while the leaves grew through the hole in the cork. The onions were never placed in water. When the roots were 1—2.5 cm. long and absolutely straight, one could be exposed to a root from another onion without being cut or even touched and without the need of glass tube holders. The exposed roots turn downwards, and a very careful investigation showed that the number of mitoses on the exposed part of the root was distinctly larger than on the opposite half, whether all mitotic stages were included or only the more conspicuous ones. The sender roots were taken as controls, and they showed a uniform and symmetrical distribution of mitoses. Almost always, the exposed root grew more rapidly than the other roots of the same onion.

When the sender root was substituted by a needle of stainless steel, the exposed root also turned downwards, but the microscopic analysis showed no increase in mitosis at the exposed side. However, the symmetrical distribution was disturbed.

The object of PAUL's investigation was the establishment of a good method for studying mitogenetic rays with onion roots. The number of examples given is not large enough to draw many more conclusions. The paper verifies GURWITSCH's principal experiment, however, and it will be the starting point for all future work with this type of detector.

b) The yeast bud method

BARON (1926) suggested that the rate of bud formation of yeast could be used as indicator of mitogenetic radiation. In his first experiments, he spread yeast over the surface of solidified nutrient agar containing glucose, allowed it to grow for from 9 to 15 hours at room temperature, and then exposed it to the radiating source for definite short periods, usually not over 30 minutes. The yeast was then incubated for from 1 to 2 hours to permit the radiation effect to develop. After this, the yeast was spread on glass slides, dried and stained. The measure was the percentage of yeast cells showing buds. When this percentage was higher in the irradiated culture than in unexposed controls, it was considered a proof of a mitogenetic effect. The original method has since been changed in some details by BARON (1930) and GURWITSCH (1932) (see p. 66).

Table 19. Effect of Yeast, of Sarcoma, and of Bone Marrow upon the Budding Intensity of Yeast

Sender	Percentage of Buds in Yeast					
	Yeast		Jensen Sarcoma		Bone Marrow	
	Control	Exposed	Control	Exposed	Control	Exposed
Experiment No. 1	24	33	21	32	30	38
2	22	30	20	35	27	32
3	13	25	20	27	21	39
4	14	25	20	26	28	38
5	24	33	28	35	29	39
6	23	33	30	38	20	35
7	25	34	25	35	23	37
8	27	35	25	36	22	34
9	25	36	25	35	27	36
10	26	36	21	30	26	33
Average	22.3	32.0	23.5	32.9	25.3	36.1

The most extensive early data with this method have been published by Siebert (1928a). Table 19 gives some of his experiments. The numbers indicate the percentages of yeast cells with buds. In all his experiments, sender and detector were separated by a quartz plate. It is now customary to record results as "induction" effect, i.e., as increase in buds of the exposed yeast over the control, expressed in percents of the control value. Thus, when the exposed yeast shows 33% buds, and the control, 24%, the increase is 9, and this is an increase of 37.5% over the control. The induction effect is 37.5%.

$$I = \frac{100\,(\text{exposed} - \text{control})}{\text{control}}$$

Siebert (1928b) used this method for a number of interesting studies in physiology. He observed the working, or excited muscle to radiate strongly while the resting, quiet muscle did not do this (Table 20). He attempted to produce radiation by changing chemically the pulp of resting muscle to that of working muscle. Finally, he succeeded by placing the acidified pulp in an oxygen atmosphere, since the addition of lactic acid alone would not produce radiation. Moreover, he obtained positive results in air by using a very dilute $CuSO_4$ solution as oxygen catalyst. When

Table 20. **Effect of Electrically Excited and of Resting Frog Muscle upon the Budding Intensity of Yeast**

Experiment No.	Percentage of buds in yeast when exposed to muscle		Experiment No.	Percentage of buds in yeast when exposed to muscle	
	Resting	Excited		Resting	Excited
1	32	45	13	23	41
2	24	32	14	22	33
3	23	36	15	23	32
4	25	36	16	21	30
5	26	41	17	22	29
6	22	30	18	22	32
7	22	31	19	22	32
8	22	28	20	23	30
9	26	35	21	25	33
10	26	34	22	25	33
11	26	35	23	24	30
12	26	31	24	24	31

he finally found that $\frac{N}{10000}$ KCN solution would prevent radiation, he concluded that the source of radiation must be chemical. Thus started the first experiments about chemical reactions as the source of radiant energy (p. **33**).

Siebert later (1930) concentrated his attention upon blood radiation. He verified the statement of Lydia Gurwitsch and Salkind (1929) that blood of normal, healthy people radiated distinctly, while that of cancer patients did not. He found, further, that urine radiated, and that there was a good parallelism between blood and urine radiation. Of 35 patients with cancer, the majority showed no radiation of blood or urine. The exceptions were patients after recent treatment with X-rays and isamine blue.

Anemia, leucemia, high fever (sepsis, pneumonia, scarlatina) prevented radiation of blood, as well as of urine. With syphilis, radiation varied, but blood and urine went parallel. None of the other diseases tested caused loss of blood radiation (details see p. **153**).

The experiments on the metamorphosis of amphibia and insects (p. **167**), and on the healing of wounds in animals (p. **173**), by Blacher and his associates, were all carried out by the yeast bud method. The importance of the yeast agar blocks in the establishment of biological spectra has already been mentioned on p. 35. This method is very commonly used in mitogenetic investigations at the present time.

Method: GURWITSCH gives the following directions for the yeast bud method (1932, p. 7): Beer wort agar plates are flooded with a very fine suspension of yeast in beer wort, the liquid is distributed evenly over the agar surface by careful tilting, and the surplus liquid is drawn off with a pipette. After about 5 to 6 hours, the surface is covered with a fine, delicate film of yeast, and is now sensitive, and remains so until about the twelfth hour. — Neither the temperature nor the concentration of the yeast suspension nor its age is mentioned. The temperature is probably room temperature, and it should be kept in mind here that on p. 14, GURWITSCH mentions 12° C as room temperature. The directions are probably meant primarily for the yeast *Nadsonia fulvescens* which has been used most commonly by the Russian workers, though beer and wine yeasts are occasionally mentioned.

In order to give a better conception of the proper physiological condition of the detector plate, we quote from the same book of GURWITSCH's p. 317: "The most appropriate stage of the detector plate corresponds to a thickness of the yeast growth of about 25 to 30 layers of cells . . . (p. 318). We can be certain that the lowest layers of cells which are in immediate contact with the nutrient medium consist essentially of young cells in rapid multiplication . . . The cells of the middle layers are not in optimal condition, and are no more capable of developing spontaneously the maximal energy for development which was found in the lowest layers . . . It can hardly be far from the truth to deny any appreciable multiplication in the topmost layers. However, they are not real resting forms as yet."

The method of making smears to count the buds is not given in GURWITSCH's book in any detail. It consists simply in smearing the yeast cells on a glass slide, drying and staining them. Only those buds are counted which are smaller than half of the full-grown cell. Some authors limit their counts to even smaller buds. GURWITSCH recommends that the person counting the buds should not know which slide or experiment he has under the microscope; this prevents subconscious arbitrary decisions.

Attention should be called here to a leaflet published by Ing. G. TERZANO & C., Milano, manufacturers of the "hemoradiometer" of Protti's. All conditions are quite precisely standardized, and this may account for the good results of Italian investigators.

This method has been varied by other authors. TUTHILL and RAHN (1933) studied the mode of bud formation of Burgundy yeast on raisin agar at 30° C. A typical result including all sizes of buds is shown in fig. 32. The culture immediately after being transferred contains but very few buds, and new buds are not formed at once. The old yeast cells which had ceased to multiply require some time before their reproductive mechanism is working normally. During this rejuvenation process, commonly called the lag phase, cells respond most promptly to mitogenetic rays. It is also seen that they soon reach a maximum percentage of buds. If, at this latter stage, the rate of cell division were accelerated by mitogenetic rays, it could not increase the percentage of buds (see also p. 69). The most opportune time for using such a plate as detector is evidently about an hour or two before bud formation begins. If such a detector is exposed for 30 minutes, and then incubated for one hour (so that the mitogenetic effect might manifest itself by an increase in buds) we should have bud formation beginning on the exposed plate while the control has not yet become quite ready for budding.

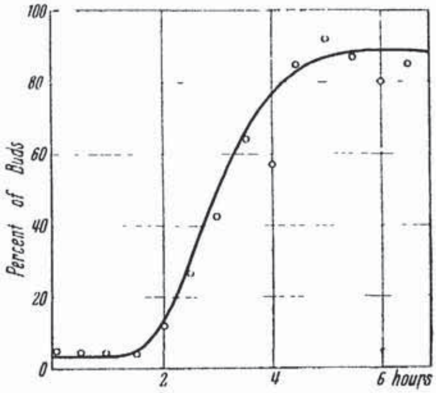

Figure 32. The development of buds in an agar surface culture of wine yeast.

The "lag period", i.e. the time interval between the seeding of the plate and the first active bud formation, depends upon the age of the culture used for the seeding. With the Burgundy yeast employed by TUTHILL and RAHN, a plate seeded with a 24 hours old culture was a good detector immediately after seeding When a 6 days old culture was used, it was advisable to incubate the plate for about 2 hours before exposure (see Table 21), or to incubate for several hours after exposure if exposure had taken place immediately after seeding.

In liquid cultures, the old yeast cells retained their buds for many days while old cultures on agar surface lost them readily. Since a low initial percentage of buds is very desirable, inoculation with yeast from agar surface cultures is recommended.

This detector is really quite different from BARON's or GURWITSCH's described above. All yeast cells are at the same stage

Table 21. **Effect of Irradiating Yeast Surface Cultures after Incubation for Different Times**

Length of Exposure: 30 minutes
Incubation after Exposure: 30 minutes

Age of Parent Culture		Age of culture when exposed					
		0 hrs	0.5 hrs	1 hr	1.5 hrs	2 hrs	2.5 hrs
		Percentage of Buds					
24 hours. .	exposed area	25.0	29.2	4.6	4.2	9.8	16.4
	control area	3.6	3.5	4.4	4.6	9.5	18.4
	increase	**21.4**	**25.7**	0.2	—0.4	0.3	—2.0
	Induction effect	**595.0**	**724.0**	5.0	—1.0	4.0	—11.0
6 days. . .	exposed area	7.0	6.0	8.5	22.5	26.2	44.4
	control area	6.0	7.0	7.6	19.0	17.0	27.8
	increase	1.0	—1.0	0.9	3.2	**9.2**	**16.6**
	Induction effect	17.0	—17.0	12.0	17.0	**54.0**	**62.0**

of earliest rejuvenation when exposed, and the cells are far enough apart not to influence each other. The "mitogenetic effect" is much greater than in the other method because there are no old, inactive cells to "dilute" the counts. In fact, these are probably the largest mitogenetic effects ever recorded. It is quite permissable to count all buds because the percentage at the beginning is very low, and a limitation in size is not necessary. This should make the counting easier.

On the other hand, this type of detector is so different from the BARON type that it may react differently in certain experiments. As long as they are used merely as detectors to prove the existence of radiations, both types are good. Probably, with very weak radiations, the BARON type is more sensitive because the old cells act as "amplifiers" (see p. 127).

Method by TUTHILL and RAHN (designed for Burgundy yeast): The yeast is kept throughout the experiment at 30° C. A 24 hours old culture in raisin extract[1]) is flooded over a solidified, sterile raisin agar

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[1]) Raisin extract: 1 pound of chopped, or seeded raisins is heated with 1 liter of water in steam for 45 minutes, the extract is pressed off, made up to 1 liter, 5 g. KH_2PO_4 and 5 g. yeast extract (or meat extract) are added; the resulting medium is sterilized at 100° C. The pH is about 4 to 4.5.

Raisin agar: Melted 6% water agar is mixed with an equal volume of the above raisin extract and sterilized by heating for 20 minutes at plate; the surplus liquid is poured off, and the culture permitted to develop for 24 hours. This surface growth is washed off with 5 cc. of sterile water, the suspension is then diluted 1 : 100 with sterile water, and with this dilution, some sterile solidified plates of raisin agar are flooded, the surplus liquid is drained off at once, and the plates should be exposed within half an hour. Half of the plate should be shaded to serve as control.

The length of exposure will depend upon the intensity of the sender: 30 minutes proved a good time with young yeast cultures. One to two hours incubation, counted from the beginning of the exposure, was the most suitable time to bring out the differences.

In these plates, the yeast cells are so far apart that the buds can be counted directly on the agar surface. The organisms are killed by placing a cotton wad with tincture of iodine in the Petri dish. Soon after that, a coverglass can be placed on the agar surface, and the slightly-stained yeast is observed in situ, eliminating all possibility of breaking off buds by smearing on glass.

In all methods where growing cells in glass or quartz containers are used as detectors, radiation from these growing cells may be reflected by the glass or quartz walls, and may thus produce radiation effects in controls as well as in the exposed cultures. Protection against reflection is advisable in all such cases (see p. 80).

Liquid cultures have also been used successfully. Here, too, the rule applies, that increases in the bud percentage can be expected only during the lag phase (see fig. 32). When all cells are out of the lag period, and produce buds at a constant rate, the percentage of buds cannot be changed by a change in the growth rate. Since this has been overlooked by some experimentors, e.g. by RICHARDS and TAYLOR (1932), it may be advisable to explain this important point in more detail.

Let us, for this discussion, distinguish five equal periods in the complete cell division of the yeast, 4 with buds and one without. Fig. 33 shows a first approximation of a "cross section" through a yeast population growing at a constant rate. It requires 5 time units for each cell to complete the cycle, i.e. to produce two cells of the same developmental stage. The percentage of buds is not constant, but fluctuates between 67 and 80%. This fluctuation is due partly to the arbitrary selection of 5 stages, but mostly to an error in the cross section. In a growing culture, there must

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100° C. On account of the high acidity, the agar becomes hydrolyzed under pressure, and fails to solidify; the same is true with prolonged heating at 100°.

necessarily be more cells of the young stages than of the old. If we take all cells of the first 5 units as a more appropriate cross section, we obtain the following picture:

Yeast Culture at a Constant Growth Rate

Time units	0	1	2	3	4	5	6
	9 no buds	9 tiny	9 small	9 medium	9 large	18 no buds	18 tiny
	8 tiny	8 small	8 medium	8 large	16 no buds	16 tiny	16 small
	7 small	7 medium	7 large	14 no buds	14 tiny	14 small	14 mediu
	6 medium	6 large	12 no buds	12 tiny	12 small	12 medium	12 large
	5 large	10 no buds	10 tiny	10 small	10 medium	10 large	20 no bud
with buds.	26	30	34	39	45	52	60
without...	9	10	12	14	16	18	20
Total.....	35	40	46	53	61	70	80
% buds. .	74.4 %	75.0 %	73.9 %	73.7 %	73.9 %	74.4 %	75.0 %

There is a fluctuation of only about 1%.

This percentage depends only upon the variety of yeast used. Whether it grows rapidly or slowly, whether the time unit is 30 to 60 minutes (at cellar temperatures) or 10 minutes (under optimal conditions), the percentage of buds remains 74—75%. A change of the growth rate would not affect the bud percentage at all.

However, a change would become noticeable if only one certain stage of the cycle should be accelerated. If mitogenetic radiation should speed up the very first stage so much that the "tiny" buds never appeared, the number of buds at the 0 period would drop from 26 out of 35 to 18 out of 27, or from 74.4 to 66.8%, and this lower level would continue as long as radiation accelerated the one particular stage.

It is rather probable that only a certain stage of the cell cycle is affected by mitogenetic rays. Many observations suggest this, and our present conceptions of the mechanism of cell division do not contradict it. This would offer a good explanation for the "false mitogenetic depression" of GURWITSCH (1932, p. 210) and especially of SALKIND (1933) where the percentage of buds decreases while the total cell count increases under the stimulation of mitogenetic radiation.

The reliability of the BARON method has been doubted by NAKAIDZUMI and SCHREIBER (1931) who claimed to have followed BARON's method explicitly. However, they have kept their detector cultures for 9—12 hours at 25° C while BARON used room temperature which may be as low as 13° C in Russia. That their cultures were far too old, can be seen from the fact that in most of their experiments, the percentage of buds decreased distinctly in 2.5 to 8 hours.

The claim of these authors that the error of the method has never been considered by the workers is entirely wrong. Every biologist knows the error of his methods, at least approximately. The Russian investigators have repeatedly stated the error of their method, though they have not given all the data necessary for others to check their computation. This can be done very easily, however, from the data on experiments without mitogenetic effect. SIEBERT e. g. rarely publishes less than 10 experiments to prove an yone point. The following computation is made from a study of the effect of KCN upon organic catalysts (1928b and 1929), and the differences between control and poisoned catalyst give the error which is computed here in two different ways:

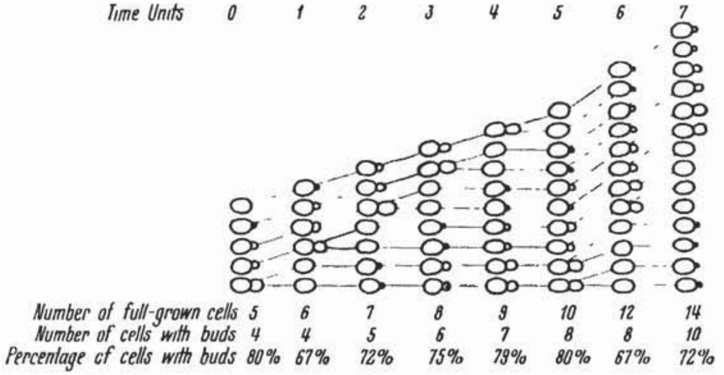
Time Units 0 1 2 3 4 5 6 7
Number of full-grown cells 5 6 7 8 9 10 12 14
Number of cells with buds 4 4 5 6 7 8 8 10
Percentage of cells with buds 80% 67% 72% 75% 78% 80% 67% 72%

Figure 33. A schematic representation of the bud formation of a yeast culture growing at a constant growth rate.

	average mean deviation	average increase through radiation	standard deviation %	increase by radiation %
1928 a Table VI	1.2	9.2	3.3	35
VII	1.4	7.9	2.9	28
VIII	2.3	10.9	4.6	35
1929 Table I	1.1	13.4	2.6	67

The increases resulting from irradiation are very much larger than the error, regardless by what method it is computed.

e) Detection by increase in cell number

To the investigator not familiar with yeasts, it may seem that there should be no essential difference between the total number of cells or the percentage of buds as a measure of the growth rate. For this reason, the preceding pages were written. There is a difference so fundamental that in a number of experiments, the yeast bud method indicates a decrease in the growth rate while the total cell count shows an increase (e. g. Table 34 p. 116).

The number of cells is the best measure of the growth rate, better than any other of the methods described in this book, because it ascertains cell divisions directly while the mitoses in onion roots, the buds of yeast cells, the respiration, etc. are indirect measurements of growth, and may be influenced by secondary effects. This method is used with yeasts as well as with bacteria. While they are identical in principle, they differ in the method of measuring since yeasts are so much larger than bacteria; they shall be treated separately in this chapter on Methods.

(1) Yeasts: The number of yeast cells in a liquid may be ascertained by plate count, by hemacytometer count, by measuring the volume of all cells, or by comparing the turbidity by means of a nephelometer.

The plate count method has rarely been used. It is the same as that used for counting bacteria except that more preferable media are beer wort agar or raisin agar.

The hemacytometer has been used by the Russian workers as well as by HEINEMANN (1932) in his studies on blood radiation.

HEINEMANN's method for testing the radiation of blood consists essentially in the exposure of a 12-hour culture of beer yeast (temperature not mentioned) in liquid beer wort, to the radiation of blood diluted with an equal amount of a 4% $MgSO_4$ solution to prevent coagulation. The exposure lasts 5 minutes, and it is made discontinuous by moving some object between blood and yeast at intervals of 2 seconds (see p. 103). 0.5 cc. of the exposed yeast suspension is then mixed with an equal amount of beer wort and incubated for three hours at 24° C. The total number of yeast cells (counting each smallest bud as an individual) is determined with a hemacytometer at the start, and again after three hours' incubation, both in the control and in the exposed culture. All cultures are preserved for counting by the addition of H_2SO_4. The control is counted twice. An example of the results and of the method of calculation is given in Table 22.

Table 22. Yeast Cells Counted in Hemacytometer *before* and *after* Irradiation by Blood

	start	Control after 3 hours		Irradiated	Increase of		,,Induktion" – $\frac{100\,(\text{Irr.—Con.})}{\text{Control}}$	actual increase in growth rate
		a	b		Control %	Irradiated %		%
Healthy person .	41.5	62	63.0	84	49	102	+108	82
Carcinoma patient	46.0	68	68.5	56	48	22	—54	—42
Chronic tonsilitis.	29.0	48	48.5	49	65	65	+ 0	0

Heinemann emphasizes that this method depends upon the physiological condition of the yeast, and that no effect can be expected when the control grows too rapidly, i. e. when the control increases to more than double during the incubation period. This means, in other words, that the yeast must be in lag phase, else it would double in less than 3 hours. To avoid errors from this source, he tested each blood sample with two different yeast strains

The results by this method verified all former experiences with blood radiation, especially in regard to cancer patients, as will be shown in Chapter VII. He also added some important new facts regarding radiation of the blood of old people and of patients with chronic tonsilitis.

The measurement of the growth rate of yeast by cell volume has been studied in detail by Lucas (1924). The accuracy is not greater than with plate counts, and the curves and data published by this author show so little deviation only because they are presented in logarithms instead of actual numbers. However, the volume is sufficiently accurate to prove mitogenetic radiation, and the method requires less time and less eye strain than the hemacytometer method, also giving quicker results than the plating method. Brainess (quoted from Gurwitsch, 1932, p. 17) has adapted the method for the small volumina available in mitogenetic work.

Volumetric Method: Kalendaroff (1932) used beer yeast in a strong wort (18—22° Balling); only cultures 15—20 hours old (temperature not given) which are actively fermenting, are suitable as detectors. After exposure, the yeast is distributed evenly, and a definite amount of the exposed culture is measured by means of a micropipette, e. g. 0.2 cc. This is added to 1 cc. of fresh wort, and incubated for 4 hours at 28° C. The

Table 23. Height of yeast column of centrifuged yeast cultures, exposed to the various spectral regions of the radiation produced by gastric digestion of serum albumin, and of their controls

Wave Length	mm of yeast column		Induction Effect	Wave Length	mm of yeast column		Induction Effect
	control	exposed			control	exposed	
2320	15	15	0	2360 -	18	17	0
2330	15	15	0	2370	17	18	0
	15	15	0				
	14	14	0				
2330 -	20	23	15	2380	12	16	33
2350	20	24	20	2390	9	12	33
	20	25	25		9	12	33
	20	26	30				
	20	25	25				
	20	26	30	2390-	15	18	20
				2400	15	20	33
2350 --	19	19	0		21	26	24
2360	19	19	0		20	24	20

yeast cells are killed by adding 0.2 cc. of 20% H_2SO_4, and are centrifuged in pipettes commonly used for measuring the volume of blood corpuscles (the illustrations of the Russian workers appear to be VAN ALLEN hematocrit tubes). The yeast column of the exposed sample is compared with that of the control.

Table 23 shows some results obtained with this method by BILLIG, KANNEGIESSER and SOLOWJEFF (1932) who used it in the determination of the spectrum of gastric digestion.

A still more rapid method for estimating the amount of growth is the measurement of the turbidity of the culture by means of a nephelometer. This method has been used occasionally by bacteriologists for several decades; the methods are reviewed and analyzed by STRAUSS (1929). The nephelometer can be used for bacteria as well as for yeasts, while the cell volume of bacteria is too small to be measured with sufficient accuracy in the earlier stages of growth. Attention may be called to the description of a simple nephelometer by RICHARDS and JAHN (1933). More complicated is the differential photoelectric nephelometer described by GURWITSCH (1932, p. 17).

Table 24. *Bacillus mesentericus* Irradiated Continuously by Yeast at 12 mm. Distance, through Quartz

Time	Cells per cubic millimeter			Mitogenetic Effect
	start	control	irradiated	
2 hrs	3168	11 440	18 290	60
2.5 ,,	528	7 920	8 976	13
3 ,,	4048	12 496	16 016	28
3.5 ,,	1584	10 912	13 552	24

Time	Generation Times control	irradiated	increase
	min.	min.	%
2 hrs	63.7	46.8	36
2.5 ,,	38.1	37.7	2
3 ,,	110.8	90.7	22
3.5 ,,	75.5	67.8	11

Another way of estimating the amount of growth in yeast cultures has been suggested by BARON (1930) who compared the size of yeast colonies in hanging drops. This method has been slightly modified by BORODIN (1934) who photographed the colonies and measured their area with a planimeter.

(2) Bacteria: The stimulation of bacterial growth by mitogenetic radiation had already been observed by BARON (1926) and by SEWERTZOWA in 1929. Table 24 gives some of the results obtained by the latter. The data were verified by ACS (1931), who irradiated liquid cultures of *Bacillus murimors* with agar cultures, either of the same species, or of yeast, and found that the effect by "muto-induction", i. e., by the same species, was the greater.

BARON and ACS did not recommend the growth rate of bacteria as a universal indicator for mitogenetic radiation. This was done most successfully by WOLFF and RAS (1931) These authors worked with different species, and the number of cells was determined by the customary method of bacteriological technique, the agar plate count Instead of making dilutions in water, they took their samples with a WRIGHT pipette which delivers $\frac{1}{250}$ of a cc. using the slide cell method. FERGUSON and RAHN (1933) obtained good results with the $\frac{1}{100}$ cc. pipettes used in the standard (Breed) method for the microscopic count of bacteria in milk.

The plating of such minute quantities is necessary because only very small amounts of the culture (about 1 cc.) can be exposed, on account of the strong absorption of ultra-violet light by the customary bacteriological media. WOLFF and RAS (1931) showed that a layer of standard nutrient broth 0.5 mm. thick transmitted only rays above 2500 Å; if broth is diluted with 9 parts of water, a layer of 1 mm. still transmits some rays as low as 2200 Å. WOLFF and RAS irradiated their bacteria in standard broth in a layer of 0.6 mm.; even then part of the bacteria were shaded.

FERGUSON and RAHN (1933) verified this observation. 1 cc. of a standard broth culture of Bacterium coli in a quartz dish in a layer of 0.6 mm. irradiated from below showed no increase over the control, while a culture in broth diluted 1 : 10 showed a good mitogenetic effect. WOLFF and RAS pointed out that only during the lag phase, definite results could be obtained. During rapid growth, there was no effect.

A most interesting observation was the exhaustion of bacteria by continued irradiation, resulting in a decreased growth rate. The two experiments in Table 25 show a lag period of about 2 hours in the control Irradiation decreases this period very distinctly, and with increasing intensity, or decreasing distance, the lag becomes shorter and shorter. However, continued irradiation after the lag phase retards the growth for some time, and at 5 hours, the control shows more cells per cc. than any of the cultures whose growth was distinctly stimulated. This retardation of growth is only temporary; most of the irradiated cultures almost doubled their number during the 5th hour, indicating a return to the normal growth rate (fig. 34).

Method (the most recent method by WOLFF and RAS, 1933a). A fresh suspension of staphylococci in broth, with about 20 000 cells per cc., is placed in a glass dish in a very thin layer, covered with quartz, and exposed (e. g. to milk + rennet in a quartz tube). After exposure, the dish with the bacteria is incubated at 37° C for 15 to 30 minutes; so is the control. By means of a capillary pipette, samples of the exposed culture and control are either plated on agar, or brought into "slide cells" according to WRIGHT.

Table 25. The Effect of Different Intensities of Continous Radiation through Quartz upon the Rate of Growth of *Staphylococcus aureus* Sender: Agar surface culture of *Staphylococcus aureus*, at various distances

Distance between sender and detector		Cells per cubic centimeter			
		Control	12.5 cm.	5 cm.	2 cm.
Experiment I	start	31 200	31 200	31 200	31 200
	after 1 hour	31 400	32 200	**39 700**	**50 200**
	,, 2 hours	32 100	**55 700**	54 000	48 800
	,, 3 ,,	45 000	57 600	51 700	46 800
	,, 4 ,,	133 000	128 500	117 000	51 200
	,, 5 ,,	262 000	237 000	135 000	79 200
Experiment II	start	14 700	14 700	14 700	14 700
	,, 1 hour	14 650	14 200	16 400	**24 500**
	,, 2 hours	15 100	16 600	**32 400**	20 250
	,, 3 ,,	17 100	**28 700**	32 700	22 500
	,, 4 ,,	50 700	80 800	46 700	21 400
	,, 5 ,,	123 000	108 500	79 300	44 400

The unirradiated controls never show growth in this short time while the irradiated cells do.

Wolff and Ras consider over-exposure the most common cause of failure: over-exposure either produces no effect at all, or eventually a decrease of growth rate (see p. 115).

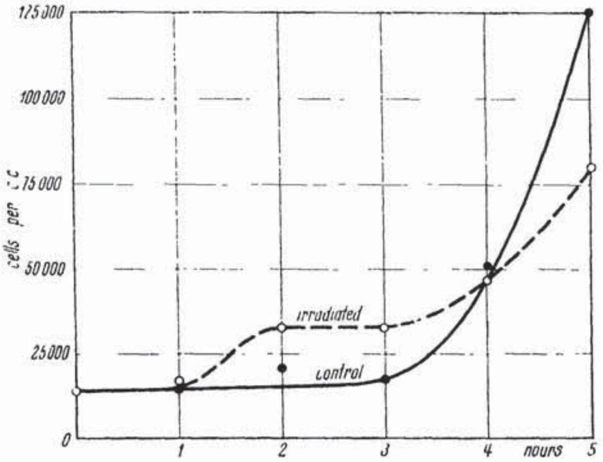

Figure 34. Developpement of two liquid staphylococcus cultures of which one was exposed continuously to the radiation from a staphylococcus agar plate.

The error of the method is mentioned in 1934; four detailed series are given, the counts being 73.1 ± 3.6; 91.2 ± 3.3; 133.8 ± 4.5 and 171.1 ± 7.3. The errors are between 3.4 and 4.9% of the total count; the increase by irradiation amounted to 25% and 28%.

Method: FERGUSON and RAHN (1933) studied the best conditions for a good mitogenetic effect with *Bacterium coli*, and found that it depended primarily upon the age of the culture, and the transmission of ultraviolet by the medium. 24 hours old cultures never reacted; cultures 48 hours old or still older always responded. The best medium was 1 part standard broth plus 9 parts water. The simplest procedure is to irradiate 1 cc. of an old culture in dilute broth (either in a quartz dish of 4—5 cm. diameter, through the bottom, or in a quartz-covered glass dish, from above), dilute this 1 cc. after exposure 1 : 10 000 with dilute broth, incubate, and plate at least every 2 hours for 6 to 8 hours. The effect may not become apparent if the cell concentration is too high (over 100 000 per cc. at the start of incubation, see Table 41 p. 136). The time of exposure depends upon the intensity of the source, and no rules can be given. With a 4 hours old agar surface culture (37° C) as sender, the best results were obtained with 15 to 30 minutes of irradiation (see Table 26).

3. Computation of the Induction Effect: The induction effect in these bacterial cultures can be computed in the same manner as explained on p. 64. As a matter of fact, this has been done by SEWERTZOWA (Table 24) and ACZ. This computation implies that multiplication of bacteria is arithmetical, while in truth it is exponential. By computing the growth rates, we have a really reliable measure. The growth rate is usually substituted by the generation time, i. e. the time required for the average cell to double. It is computed from the formula

$$g = \frac{t \times \log 2}{\log b - \log a}$$

where *a* is the number of cells at the beginning, and *b* the number after the time *t*. Both methods have been applied in Table 26. The induction effect calculated from the numbers directly is in one case (15 minutes exposure) 183, while the effect in the same culture, computed from the generation time, is only 27. However, it must be considered that the number 183 has no real biological significance while the other indicates that during the four hours, this culture had a growth rate 27% higher than the average of the two controls.

Table 26. 3 days old culture of *Bacterium coli*, irradiated for various lengths of time by an agar surface culture of the same bacterium

(The numbers are cells per cc. after diluting the irradiated cultures 1 : 10 000 with broth)

	Control No. 1	Duration of Irradiation				Control No. 2
		60 min.	30 min.	15 min.	7.5 min.	
start	5 050	5 650	6 800	7 250	6 250	6 550
after 2 hours . . .	5 700	5 800	7 950	7 750	5 850	7 000
,, 3 ,, . . .	—	11 000	14 350	14 400	8 300	8 500
,, 4 ,, . . .	23 750	24 250	29 000	35 350	19 150	19 550
,, 6 ,, . . .	188 000	223 000	434 000	457 000	139 000	137 000
Induction Effect .	—	37	168	183	−16	

Generation Times,
in minutes, for the time interval from 2–6 hours

Generation Times	47.6	45.5	41.5	40.8	52.5	56.0
Induction Effect .	—	+14	+25	+27	−1	—

This procedure has also been used in Table 24. It could be used with yeasts as well, e. g. in Table 22. However, the error becomes very large if the increase is small. In the tables given by Schreiber (1933), the generation time of yeast for the first 2 hours when the mitogenetic effect is strongest is mostly more than 2 hours. This means that not all cells had divided in this time. Though Schreiber does not give the actual numbers of cells from which he calculated the generation times, it seems from his curves that they were computed from less than 100 cells. This makes the error very large, and a comparison of the growth rates must necessarily result in enormous percentual differences. Thus, Schreiber found the variations in duplicate plates of *Saccharomyces ellipsoideus* commonly to reach 40%, and occasionally more, and in one case even 163%. With *Nadsonia*, the deviation of duplicates went as high as 237%. With such a large error, there is little hope of detecting mitogenetic effects.

The "Induction Effect" as usually calculated (p. 64) has no definite meaning. It permits no comparison with the probable error of the method. It is very unfortunate, therefore, that many investigators, especially the Russian scientists, record the obtained effects merely by giving the "Induction Effect". The critics point out very justly that such relative numbers are not convincing. It would add a great deal to the general recognition of biological radiation if the actual data obtained (numbers of cells, of mitoses, percentage of buds etc.) were given for the exposed culture, the control and also for the same culture before the beginning of the experiment.

d) Detection by cell division in larger organisms

The three detectors mentioned above are the only ones that have been commonly used to prove the existence of mitogenetic radiation, a few others have been employed occasionally, but not often.

Mention is made of the reaction of mold spores upon mitogenetic rays. The first publication of actual effects is probably that by SCHOUTEN (1933). WOLFF and RAS (1933c) mention that they react slowly and require about ten times as long an exposure as staphylococcus cultures.

FERGUSON and RAHN, in some unpublished experiments, observed that the radiation of the detector culture may be reflected, and may thus give a mitogenetic effect even in the controls which received no radiation from outside. Spores of *Aspergillus niger* were spread on an agar surface. When the dish was covered with a glass cover or a quartz plate, germination was more rapid than when the cover consisted of black paper or sterile agar. The effect varies in magnitude, and is not always present, but must be guarded against in this technique and probably in most others. This reflection may be the cause of many failures to observe mitogenetic rays. The strong effect can be explained by polarisation of the rays through reflection (see p. 45).

Exper. No.	Percentage of germinating mold spores			
	Reflecting surface		Non-reflecting surface	
	Glass	Quartz	Agar	Black paper
13	36.0	35.0	19.8	19.6
14	33.3	35.1	14.5	23.3
15	42.8	39.0	22.4	35.2
16	33.2	—	—	37.4
17	—	45.8	18.4	14.8

In the animal kingdom, the eggs of the smaller animals have been used occasionally to demonstrate the mitogenetic effect. **Reiter** and **Gabor** (1928) showed that frog eggs when irradiated with the spectral line 3340 Å developped more rapidly into tadpoles than the controls. Too long an exposure retarded the development. The wave length is unusual as in all publications by **Reiter** and **Gabor** (see p. 60).

In a short paper, **Wolff** and **Ras** (1934b) showed that eggs of the fruit fly *Drosophila melanogaster* hatch more rapidly after having been exposed to the radiation from bacterial cultures (see also p. 144).

The eggs of sea urchins were found to be quite good detectors. The rate with which they divide, can be easily seen under the microscope, and the percentage of eggs in each of the different stages is a good indication of the growth rate. **Salkind**, **Potozky** and **Zoglina** (1930) were the first to show that biological radiation from growing yeast or contracting muscle will increase the rate of development of the eggs.

The Italian school of mitogeneticists has also used sea urchins repeatedly. Not all species are equally well adapted as detectors, some being much more sensitive than others. Table 27 shows some data by **Zirpolo** (1930).

Table 27. Mitogenetic Effect upon the Eggs of the Sea Urchin *Paracentrotus lividus*

Sender	Percentage of Eggs					
	Control			Irradiated		
	unchan.	2 blast.	4 blast.	unchan.	2 blast.	4 blast.
Bacillus Pierantonii	82.6	13.9	3.5	10.7	87.6	1.7
,, ,,	74.5	25.5	0	35.0	65.0	0
,, ,,	70.0	30.0	0	8.4	88.4	3.2
Penicillium (in dark)	97.8	2.2	0	7.9	91.5	0.6
,, (diffuse light)	70.4	25.7	3.9			

The morphological changes of the larvae brought about by irradiation of sea urchin eggs will be discussed later (p. 164) since a different principle is involved.

Tissue cultures would appear to be an interesting subject for the study of this radiation. The first investigation was started without the knowledge of **Gurwitsch**'s discovery.

Guillery (1928) observed during some experiments on the growth-promoting agents for tissue cultures, that two or three cultures in the same dish influenced one another. While the most rapidly-growing culture usually maintained its growth rate, that of the more slowly-growing ones was distinctly increased. Fig. 35 shows the relative daily increase of three cultures from the heart of chicken embryos, which were transplanted at different ages of the embryo, and therefore had different characteristic growth rates. These remained constant as long as the cultures were grown in separate dishes, but were changed when all three were continued in the same dish. Incisions in the solid medium which separated the cultures and prevented diffusion from one to the other did not prevent the mutual stimulation. Even when a strip of solid medium was completely removed between two cultures, the effect sometimes continued. When a glass slide was used to separate the cultures, the influence ceased, even if the slide did not touch the bottom, and permitted diffusion. Further experiments showed that the effect spreads rectilinearly; by placing several cultures in the same dish, and inserting small glass strips between some of them, the effect of the glass was found to be that of shading. This can be explained only as radiant energy which stimulates growth. It could also be shown that the radiation was reflected from metal mirrors.

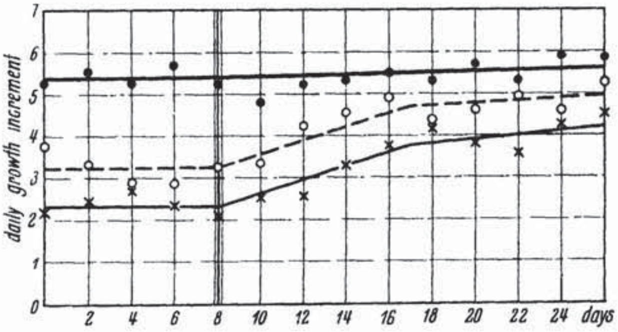

Figure 35. Daily growth increments of three cultures of chicken embryo, grown separately for 8 days, then united in the same dish.

These observations suggested to Guillery the possibility that the embryo extract which is necessary for the growth of tissue cultures, acts essentially as a source of radiation. He irradiated a number of cultures in a dish, from one side, with embryo extract, but the result was negative. However, with a head of a chicken embryo, he obtained in all 4 experiments a more rapid growth in the culture nearest to the radiating substance.

The second paper on this subject was that of CHRUSTSCHOFF (1930) who observed that growing tissue cultures began to radiate some little time after transplantation from the original tissue. With a spleen culture of *Ambystoma tigrinum*, radiation began after 60 hours; with a fibroblast culture from the heart of a chicken embryo, after 12 hours. CHRUSTSCHOFF believes that radiation is due to autolysis of necrotic parts in the culture.

Next, CHRUSTSCHOFF used the tissue culture as detector. A fibroblast culture (chicken) was divided into halves, both were cultivated in separate drops on the same quartz cover glass, and one of the two was irradiated for 48 hours by a beating embryo heart, which was replaced when necessary. At this time, the irradiated culture appeared much denser than the control; after 3 days (the last day being without radiation), the exposed culture was far in advance of the control.

JAEGER (1930) observed that blood radiation retarded the growth of tissue cultures. Here, as with all other detectors, some investigators obtained negative results. LASNITZKI and KLEE-RAWIDOWICZ (1931) as well as DOLJANSKI (1932) could find no stimulation by mitogenetic radiation. DOLJANSKI used cultures which reacted promptly upon addition of embryo extract, but they did not respond at all to organic radiations, even if the distance was only ½ mm.

Very recently, JULIUS (1935) obtained definite growth stimulation of chick fibroblast cultures, but only on poor media. By placing one half of a culture on glass, the other half on quartz, and exposing both to radiation from staphylococcus culture, those on quartz grew more rapidly as measured by means of a planimeter. The induction effect was recorded as the ratio of increase in the quartz culture over that in the glass culture. In 56 such pairs, the average effect was 1.92 ± 0.15. Another set of 48 pairs, but without irradiation, gave the ratio 1.01 ± 0.08, proving no chemical effect from glass or quartz. The actual stimulation by radiation from bacteria was therefore 0.91 ± 0.17, the effect being 5.3 times the probable error.

It may be that a certain stage of development is necessary to make the cells sensitive to the mitogenetic stimulus, as was shown for yeast and bacterial cultures (pp. 61) and 120).

Table 28. Mitogenetic Effects produced in the Corneal Epithelium of vertebrates by 3—4 minutes exposure of the left eye to the spectral line 2030 Å

Number of Mitoses								
Triton			Frog			Rat		
left eye exposed	right eye control	% incr.	left eye exposed	right eye control	% incr.	left eye exposed	right eye control	% inc
54	27	100	277	108	158	3086	1944	7
134	66	100	3200	2070	50	3312	2050	6
130	51	150	615	333	85	1644	1187	4
50	30	68	695	205	240	440 1)	196	12
53	38	40	885	221	300	2593 1)	1578	7

A very good detector is the corneal epithelium of vertebrates, according to LYDIA GURWITSCH and ANIKIN (1928). It is the only easily accessible tissue of the grown animal showing frequent mitosis. The number of mitoses varies with the physiological state; it increases rapidly with good nourishment, dropping in rats during starvation from approximately 2000 to about 50. Fortunately, the two corneae of the same animal always show very nearly the same number of mitoses, so that one eye can be irradiated, and the other used as control.

Method: For physical light sources, an exposure of 3—4 minutes was sufficient, the animal's head being held in the hands of the experimentor. With biological sources, exposure had to be continued for 20 minutes, which necessitated the tying down of the head into an immovable position. Ordinarily, after irradiation, 3—4 hours time was given for the manifestation of the effect. The cornea was fixed for 40 minutes in 70% alcohol + 5% acetic acid, stained with hämalaun, and clarified in glycerol.

The results in Table 28 show very strong mitogenetic effects.

e) Detection by changes in yeast metabolism

GESENIUS (1930a) concluded that such decisive changes as the acceleration of the growth rate of yeast must be accompanied, or perhaps preceded by changes in metabolism. He studied, therefore, the influence of irradiation upon the rate of respiration and of fermentation of yeast. The technique employed was

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1) The source of radiation was a yeast culture.

essentially that of Warburg (1923), or of Runnström (1928) for measuring respiration of tissues or tissue pulps. The organism used as detector was a wine yeast, which was exposed in quartz-bottomed dishes to yeast radiation for 4 hours before being tested. The result was a stimulation of fermentation, but a retardation of the oxygen uptake.

The results can be briefly summarized in the following way:

Fermentation in N_2—CO_2 atmosphere	102 experiments	68 increase 4 decrease 30 within the limits of error
Respiration (oxygen-uptake) in O_2 atmosphere, without sugar	54 experiments	40 decrease 1 increase 13 within the limits of error

The number of yeast cells in these tests was very large, 7 to 10 billion cells per cc. This is 10 to 100 times the maximal population which can develop in the medium used. The mutual irradiation of the cells must play an important role in these experiments, and probably accounts for the depression of respiration.

Gesenius tried further the influence of radiation upon macerated yeast, free from cells, i. e. upon zymase. While yeast radiation produced no effect, blood radiation decreased the fermentation in 28 out of 30 experiments, the average depression being 14%. The same retardation of respiration also could be obtained with sea urchin eggs during the early stages of cell division.

Gesenius (1930b) applied this test, after the improvement of the technique, to blood radiation and found that normal blood always radiated1). He observed further that from patients with most diseases, the blood radiated, and that the consistent exceptions were only with cases of pernicious anemia, leucemia, carcinoma and severe sepsis (see fig. 46 p. 153). His results agree very well with those of L. Gurwitsch and Salkind and of Siebert. The role in cancer diagnosis of this loss of radiation will be discussed in Chapter VII.

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1) "Healthy blood never fails. If a failure occurs, it is time to test either the yeast or the apparatus." (Gesenius, 1932.)

f) Morphological changes by biological radiation

Quite different from the previously described manifestations is a decided morphological change in the irradiated organisms. The first observations of this kind were those by J. and M. Magrou (1928) who exposed the eggs of the sea urchin *Paracentrotus lividus* to radiation from bacteria, yeasts or other organisms, and even from chemical reactions. They obtained quite abnormal, more or less spherical larvae while the normal larvae possess a very characteristic conical form (see fig. 49 p. 165). Upon various criticisms, the greatest care was taken in later experiments (1931) to prevent any chemical influences. In each case, the effect was transmitted through quartz, but not through glass. Table 29 shows a summary of the results of these experiments, and the purely physical nature of the effect cannot possibly be doubted.

While the Magrous originally explained this morphological change through mitogenetic rays, later experiments, together with Reiss (1931), offered another possible explanation. They found that though a layer of glass prevents the effect, two layers of glass with a coat of paraffine between them permit the effect to pass. From this and similar experiments, these authors conclude that they are dealing with an electric effect. The larvae become abnormal if the source of radiation is separated from the medium of the sea urchin eggs by a very good electric insulator, and if further the difference in oxidation-reduction potential between the two liquids is very great. When the electric insulation was prevented by a metallic connection, the larvae remain normal. This explanation is tentative. In their recent publications, the Magrous do not give it preference to the ultraviolet radiation theory.

In 1929, Christiansen observed very strange morphological changes in yeasts and in bacteria brought about by menstrual blood. The effect passed through quartz coverslips, and must therefore be considered as the result of biological radiation. The yeast cells either became large and spherical, with enormously distended vacuoles; or, they elongated and produced hyphae; or, they did not grow at all, but died.

During the last two years, the author and his associates have regularly observed similar morphological changes.

Table 29

Magrou's Results with Biological Irradiation of Sea Urchin Larvae

No. of Experiments	Source of Radiation	Larvae Separated from Source of Radiation by	% of Experiments Showing Abnormal Larvae
76	none	glass	0
4	dead bacteria	glass	0
76	*Pseudomonas tumefaciens*	quartz	75
7	same	glass	43
36	none	glass	0
25	*Staphylococcus aureus*	quartz	75
3	same, agglutinated	quartz	0
16	none	glass	0
12	*Streptococcus lactis*	quartz	93
2	same	glass	0
4	bacteria-free serum of Streptococcus culture	quartz	25
7	none	glass	0
5	Saccharomyces and Debaryomyces	quartz	100
30	none	glass	0
18	Berthelot's culture medium	quartz	100
6	same	glass	0
56	none	glass	0
36	glucose oxidized by ferricyanide, permanganate or bichromate	quartz	78
14	same	glass	0

Summary

221	none		0
118	microorganisms	glass	33
9		quartz	78
54	chemical reactions	quartz	85
20		glass	0

The effects in beer and wine yeasts produced by saliva were essentially identical with those observed by Christiansen. When irradiated by plants, however, the tendency was not a shortening but rather a lengthening of the cells; this was so pronounced with some Mycodermas that their growth strongly resembled that of mold mycelium. However, we could never observe the true branching of cells which Christiansen has described. Of the various parts of plants, the roots, young seeds, seedlings and pollen were the most effective, while leaves had little or no effect (see fig. 48 p. 162, and Chapter VII).

g) Physico-chemical detectors

Liesegang Rings: It was recognized by Stempell that a physico-chemical detector would carry much more weight than biological ones for the proof of mitogenetic radiation. He observed that the Liesegang rings are disturbed by biological radiation. These rings appear when, on a gelatin gel containing certain salts, a drop of another solution is placed causing a precipitate with the salts in the gelatin. The precipitant diffuses gradually into the gelatin, and the precipitate is deposited in concentric rings.

Method: 2 cc. of chromate gelatin (12 g. gelatin, 160 cc. water, 0,4 g. ammonium bichromate, being mixed, immediately before use, with 1 cc. water + 1 drop of 3% aqueous pyrogallic acid) are poured hot upon a clean glass plate 3.5 × 4 inches. After solidification, 2 drops of a 20% $AgNO_3$ solution are placed on the center of the plate by means of a fine pipette. Silver chromate is gradually precipitated in concentric rings wich spread over the entire plate in about 24 hours. The uniformity of these rings is disturbed by radiating material placed in quartz tubes as closely as possible over the gelatin surface.

Stempell's first experiments (1929) with onions were not accepted since it could be shown that the allyl-mustard oil of the onion caused a chemical disturbance of the Liesegang rings. In later publications, however, Stempell could prove disturbance of the rings when chemical influences were completely eliminated.

The situation is rather complicated. Strong ultraviolet light from a quartz mercury vapor lamp thrown through a narrow slit upon the gelatin intensifies the ring formation at the exposed places, while weak light, at the edge of the slit, decreases it. Onion oil acts in the opposite way; a large dose of onion oil gas lessens the ring formation while small doses intensify it.

Stempell (1932, p. 46) states that the disturbance of **Liesegang** rings is usually brought about by a combined action of radiation and chemical effect of a "gas", i. e. a volatile substance produced by the sender. He considers this chemical effect to be very important, biologically, and states that the **Liesegang** rings at present are the only detector for this substance, since none of the other detectors for mitogenetic radiation react to it.

Since this book is meant to be limited to biological radiation, the interesting speculations of **Stempell** (1932, p. 46) regarding the possible biological meaning of the chemical emanations will be omitted.

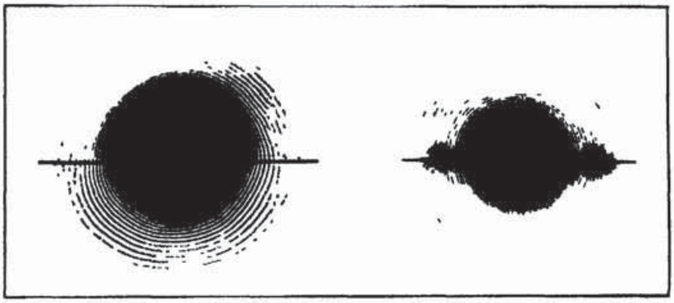

Figure 36. The effect upon **Liesegang** rings of onion base pulp in metal tubes with a slit whose position is indicated by the black line left: effect through cellophane; right: effect through 0.5 mm of quartz.

Decomposition of Hydrogen Peroxide: Another, quite different detector, has been found by **Stempell** (1932, p. 55). It is based on the deterioration of H_2O_2 into $H_2O + O$, under the influence of ultraviolet radiation. One onion root, or a bundle of several roots, is fixed so as to touch the underside of a thin quartz plate. On the opposite side of the quartz plate is placed a drop of H_2O_2, just over the root tips. After long exposure in a moist chamber, the peroxide concentration in this drop is less than that of the control.

Flocculation of Colloidal Solutions: A promising method has been worked out by **Heinemann** (1934, 1935) who observed that inorganic sols flocculate more readily when exposed to mitogenetic rays. Gold sol was found to be more satisfactory than iron hydroxides. The gold sol was prepared in the following way: To 1000 cc. of a slightly alkaline solution of 0.8% glucose, 150 cc. of a neutralized solution of 0.1 g. $AuCl_3$ are added. Upon heating, the gold salt is reduced by the glucose to a bright-red, clear gold sol. A small amount of NaCl solution is added just before the beginning of the experiment to make the gold sol unstable. This is done in complete darkness.

The solution is then distributed between two beakers, and each is covered with a quartz dish. They are placed into a very sensitive photoelectric differential nephelometer, and the supposedly radiant substance is placed into one of the quartz dishes. The difference in turbidity between the two beakers is read every minute, by means of a galvanometer. Radiation causes a more rapid flocculation, and therefore a change of the galvanometer reading while in the absence of radiation, the readings remain fairly uniform. The following readings were obtained in 15 consecutive minutes, the arrow indicating the moment when the radiating material was applied to the quartz dish:

control:	0	0	0	0	0	0	1	1.5	1.5	1	2	2	2	2	2
blood:	0 ↑	2	3	5	7	7	8	10	11	16	20	19	19	25	26
control:	0	0	0	0	0	0	1	1.5	1.5	2	2	2	2.5	3	3
NaCl, dissolving:	0	2	0	1	1	0 ↑	5	10	10	11	11	15	21		

h) Measurement by physical instruments

It may be surprising that radiation by organisms has not been recognized and proved conclusively long before this. The reason must be sought in its very weak intensity. The intensity is so slight that the most sensitive photographic plates and the most elaborate physical instruments have in most cases failed to record this radiation. The best detector is still the living organism. This is unsatisfactory since we must allow for considerable individual variation of the detector organisms, and as a rule, the measurements are not as accurate as with physical experiments.

The only photographic records are the ones reproduced in fig. 15 of REITER and GABOR's monograph, and those by BRUNETTI and MAXIA (1930) and by PROTTI (1930). None of them are absolutely convincing, and GURWITSCH has refused them all as proofs of the physical nature of mitogenetic radiation. TAYLOR and HARVEY (1932) could obtain no effect by exposing plates to frequently renewed fermenting yeast for ninety days.

Table 30. **Measurements of Mitogenetic Radiation by Means of the Geiger Counter**

Radiator	Time interval	Impacts per interval	
		Control	Exposed
	Experiments of Rajewsky		
Onion root	5 minutes	42.0±1.0	51.0±7.0
Onion base pulp . .	10 minutes	42.2±1.2	50.0±1.5
Onion base pulp . .	10 minutes	39.8±0.3	49.3±2.2
Carcinoma of mouse	10 minutes	23.4±0.6	30.3±0.5
Onion root	9 minutes	33.3±1.4	36.7±0.5
	Experiments by Frank and Rodinow		
Frog muscle . . .	6 minutes	12±1.3	40±2.6
Frog muscle . . .	8 minutes	12±1.3	20±1.7
Frog heart	11 minutes	19±1.1	23±1.2
Frog heart	5 minutes	34±2.4	46±2.8
Muscle pulp	4 minutes	13±1.8	20±2.2

In 1929, Rajewsky succeeded in obtaining direct physical proof of this radiation by means of a photo-electric counter (see p. 28). His data with onion roots and carcinoma are shown in Table 30. These experiments were repeated successfully by Frank and Rodionow (1930) with working muscle (see Table 30 and Fig. 21, p. 29). Later experiments of this nature are those by Barth (1934) and Siebert and Seffert (1934).

However, these results have not been accepted generally, at least not by physicists. Lorentz (1933, 1934) could show that bringing the radiating material near the counter, or opening a shutter between the counter and source, may definitely change the counting rate even if there is no radiation present. To enumerate: (1) If the biological material is not in a closed quartz container, the water vapor from the material, even though slight, will condense upon the quartz of the counter, changing its resistance and thus altering the counting rate. Some experimenters have even placed their moist material directly upon the quartz window which will certainly change the counting rate. (2) If the water vapor is carefully kept away from the counter, the charges which are inevitably present on the outside of the quartz tube containing the biological material are almost sure to change the counting rate. (3) In one case, at least, muscle was tetanized by means of an induction coil directly in front of the window.

Table 30a. Photo-electric yields obtained for ultraviolet light

Authors	Photoelectric material	Yield in quanta per electron	wavelength Å
Rajewsky, 1934 . .	Cd	about 1×10^2	2650
Schreiber, 1930 . .	K	„ 2×10^4	2540
Grey and Ouellet, 1933	Pt	„ 6×10^3	2540
Frank and Rodionow, 1932 . . .	Cd, Al	„ 2×10^3	2540
Lorenz 1933 . . .	Cd	„ 3×10^3	2540
Kreuchen, 1934 .	Cd, Al, Zn	„ $2—5\times10^4$	2540

To separate the effect of extremely low intensity from these other effects is very difficult even when their existence is realized. In none of the physical measurements of mitogenetic rays is it absolutely certain that these errors have been excluded, and it seems well to view with caution the positive results claimed for the physical detection experiments so far carried out.

A more careful description of the method of exposure, and a number of experiments with water blanks or non-radiant organic materials will be necessary to produce evidence which is physically irreproachable. The experiments of Siebert and Seffert (1934) who obtained increased counts with several hundred normal blood samples, but no increase with blood from carcinoma patients, are a step in that direction. Most convincing is the experiment that a counter gave a definite increase when exposed to normal blood, but showed no increase when this was removed and replaced by blood + KCN. Many more extended experiments of this general nature will be necessary to establish finally the radiant nature of the biological effects.

In a recent paper, Kreuchen and Bateman (1934) reviewed the field of physical detection and present their results in a table (see Table 30a) which gives the photoelectric yield (see p. 29) of the surfaces used by the various investigators. In all cases except the first, more than 2000 quanta are required to eject one electron. Though some early workers indicated higher sensitivities, it seems from later work that this value has never been surpassed if, in fact, it ever was reached. In his latest paper, Kreuchen (1935) obtained yields of from 10^5 to 10^4 quanta per electron from hydrogen-activated zinc and cadmium surfaces.

It would be of great advantage if some surface having a higher efficiency for the ultraviolet could be obtained. Preliminary experiments by one of the authors using magnesium surfaces sensitized by oxygen seem to offer encouraging results.

RAJEWSKY estimated from his experiments the intensity of this radiation, and found it for onion roots and for carcinoma tissue to be of the magnitude of 10^{-10} to 10^{-9} erg/cm^2/sec. (10 to 100 quanta/cm^2/sec. for the wave length 2300 Å). FRANK and RODINOW observed higher values: they obtained with pulp from muscle, with the working frog muscle and heart, values up to 2000 quanta/cm^2/sec.

For the reproduction of the various mitogenetic phenomena with physical sources of light, much larger intensities are required (about 6.6×10^5 quanta, according to STEMPELL, 1932).

i) Unaccounted failures in proving radiation

It must be stated with perfect frankness that biological detectors sometimes fail for unknown reasons. Probably all investigators working with biological detectors have been worried by such failures. Some of them have published short remarks. GOLISHEWA (1933) mentions that out of 373 experiments with blood radiation, in GURWITSCH's laboratory, 54 failed on account of a poor quality of the yeast culture which was used as detector. This cause became evident through the fact that all other associates using the same culture on the same day obtained negative results. No reason for the abnormality of the yeast culture is mentioned.

WOLFF and RAS (1933c) working with Staphylococci had a similar experience. Twice it happened that all the experiments of one day proved to be negative though the culture had reacted promptly on the previous day. It was found that the sensitivity of the culture had changed, and that a longer exposure was necessary. These authors believe that a change in the opposite direction may also take place. ACS (1932) claims to have increased sensitivity by selection.

Most of the sudden failures of cultures to react have not been published, but by discussing this point with the various investigators in this field, practically all seem to have had the same experience. Professor GURWITSCH has told the author that in his experience such a condition usually remained for several days, or even for a number of weeks, and it was impossible to produce even the simplest mitogenetic effect. Eventually the culture reacted normally again. Doctor HEINEMANN, after a very successful diagnosis of cancer by the absence of blood radiation (see p. 181) in Frankfurt and in London, with yeast as detector, suddenly experienced a complete lack of reaction, and none of the various attempts to obtain normal reactions proved successful, not even the testing of a large number of different yeast cultures. This failure induced him to look for physico-chemical methods of detection (see p. 89). Professor WERNER SIEBERT's many succesful experiments with a yeast detector have been mentioned in practically every chapter of this book. But with him, too, the yeast suddenly ceased to react, and he resorted to the GEIGER electron counter as a more dependable detector (see p. 92). The author himself has also had long periods of negative results in his laboratory, and they come and go at irregular intervals.

As a rule, the investigators do not discuss these periods of failure because there is still considerable doubt among physicists and some biologists concerning the existence of the mitogenetic phenomena, and the emphasis of such periodical failures might increase this doubt.

While this can not be denied, it does not seem wise to belittle this experience. On the contrary, by calling attention to it, it may help to explain the cause of these failures, and thereby may bring about a better understanding of mitogenetic effects. Whether it is due to disturbance by short radio waves (suggestion by GURWITSCH), to a change of sensitivity of the detector culture (WOLFF and RAS), to a retarding effect by human radiation (RAHN), to climatic changes or some other cause, is not known. The cause of this disturbance might be more readily traced and overcome by the cooperation of various laboratories. At the present, we do not know whether the culture, the experimenter or the environmental laboratory conditions have changed, in fact, it is only an assumption that the change has influenced the detector. If the cause should prove to be of such general nature as e. g., weather, cosmic rays, terrestrial magnetism, sunspots, it might be that the senders do not function under the prevailing condition.

These occasional failures have nothing to do with the error of the method. When mitogenetic effects are observed, they are outside the limits of error. The failures might be compared to the experience of expert florists that sometimes, certain plants refuse to bloom in the greenhouse. This was not caused by poor seed nor wrong soil, but remained unexplained for a long time until it was found that the number of hours of light per day decided this.

The consistent observation of this disturbance by most (if not all) investigators who have obtained large series of positive results, points out a common error in some criticisms. It has been claimed that many simultaneous parallel experiments prove more than similar experiments spread over a longer period of time. E. g. Kreuchen and Batemann (1934) state that one series of theirs is equivalent to 140 single experiments by Gurwitsch. That is a mistake. As long as it is not known why the occasional failures occur, no great stress can be laid upon the results obtained on any one day. It might be a day where the detector fails, and the multiplication of experiments on such days would only reveal the error of the method as such, but would not increase the proof or disproof of biological radiation.

B. INJURIOUS HUMAN RADIATION

It is an old "superstition" that a harmful emanation comes from the body or the hands of menstruating women. It is believed that bread dough kneaded by them will not rise, that food preserved by them will not keep, that flowers in their hands wilt readily. Experiments to prove this have been successful with some investigators (Schick, 1920; Macht and Lubin, 1927; Böhmer, 1927) and gave no results with others (Sanger, 1921; Frank, 1921; Polano and Dietl, 1924). Still, the belief in this effect seems to have been rather prevalent among medical men, and the term "menotoxin" for the hypothetical compound causing it is in general use.

Christiansen, as bacteriologist of a dairy laboratory in Germany, observed that the pure cultures used for dairy starters occasionally developed poorly and abnormally. After eliminating all other causes, it was ultimately found that this abnormality occurred during the menstrual period of the woman technician in charge of the cultures.

A detailed investigation by Christiansen (1929) led to the discovery that the effect from menstrual blood passed through quartz, and must, therefore, be considered a radiation. Aside from this proof, CHRISTIANSEN did not go into the nature of the effect. He worked with menstrual blood and saliva, but not with radiations from the body.

The blood produced either abnormal morphological changes in yeasts and bacteria, or killed them. The effect was much stronger in summer than in winter, and since one woman who had been treated with ultraviolet light in winter, continued to show strong radiation, he thought it very probable that the seasonal difference was brought about by the variation in solar irradiation. He called attention to the fact that the above-mentioned investigators obtaining positive results had made their experiments in summer, while the negative ones had been obtained in winter. Further, he showed that wine made with a pure culture yeast by a menstruating woman fermented feebly, while the control showed a vigorous normal fermentation.

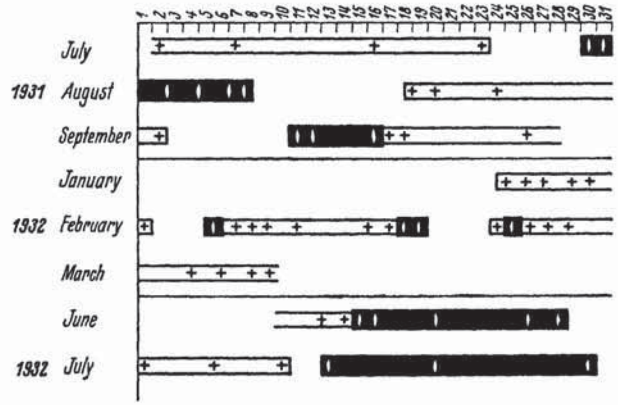

Figure 37. Alternation of growth and no growth of yeast in droplet cultures transferred by the same person.
+ signs indicate growth, o signs in black bars indicate no growth.

FERGUSON (1932) during an investigation of morphological changes in yeast by plant radiation, observed occasionally that for short periods, coverglass cultures of one yeast did not grow when made by one certain woman student. The periods of no growth alternated with those of growth through summer and fall, while in winter, the controls nearly always grew normally (fig. 37). The droplet culture on the coverglass requires that the coverglass be held in the fingers while the droplets are made with a pen dipped into the yeast suspension.

The above-mentioned experiments of CHRISTIANSEN made it probable that this failure was due to human radiation, and some experiments proved that an emanation from the fingertips of this person killed yeast in 5 minutes while others making the same test with the same culture produced no marked effects. In this experiment, the fingertip was held closely over the yeast by the support of a glass ring (fig. 38). In another experiment, a quartz plate of 2 mm. thickness was placed between finger and yeast (fig. 38); by this method, it required 15 minutes to kill the yeast. This student menstruated rarely and irregularly, and the case suggests an abnormal parallel to CHRISTIANSEN's observations.

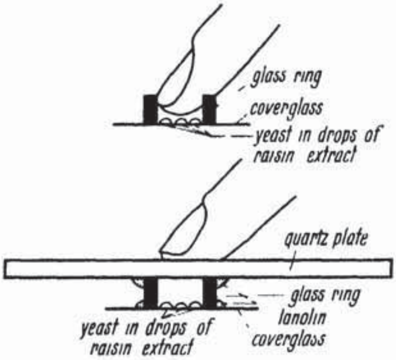
glass ring
coverglass
yeast in drops of raisin extract
quartz plate
glass ring
lanolin
coverglass
yeast in drops of raisin extract

Figure 38.
Methods of testing finger radiation.

This test has been applied to a number of people, and it was found that this radiation occurs only very rarely. It was most pronounced with a man who had recently recovered from *herpes zoster* of the face; for about 6 months after recovery, he frequently killed yeast through quartz in 15 minutes, but this was not always the case. During this time, he did not feel quite well. After a summer vacation, he felt perfectly normal, and his power of radiation was gone. With this person, radiation also occurred from the tip of the nose and from the region of the eye.1)

A third case was a hypo-thyroid patient tested only once.

A fourth case was one of the authors, during 3 successive days of sinus infection. Never before or afterwards did this person show any effect upon the yeast.

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1) This has been interpreted by imaginative but uncritical newspaper reporters as a scientific proof of the "Evil Eye", regardless of the fact that this radiation does not reach further than a few inches, and that only one especially sensitive species of yeast could be killed in this way; the authors are in no way responsible for this kind of publicity, but have not been able to prevent it.

The experiments were always made with droplet cultures through quartz, as shown in fig. 38. In all more recent work, the droplets were prepared by the same person who had been found over a 2-year period never to radiate.

The only organism which reacted upon this radiation was *Saccharomyces mycoderma punctisporus* GUILLIERMOND, isolated from the scum of a fruit juice. This yeast does not cause fermentation. Other yeasts showed slight retardation, but never was complete killing observed.

This radiation is quite likely due to a skin excretion, as will be shown later (p. 185).

Another killing effect was frequently observed with saliva. The saliva of many persons changes the oval or elliptical, granulated cells of beer and wine yeasts to spherical cells of increased size, with greatly distended vacuoles and homogeneous cell contents. Often, the organisms either do not grow at all, or cease to do so after a few cell divisions.

It is not certain, however, that the effect is one of radiation. It was observed in droplet cultures which were mounted immediately above the saliva; this did not exclude a chemical effect. When saliva was mixed with the raisin extract in which the yeast was cultivated, it produced no abnormal cells, not even with half saliva and half raisin extract. This seems to exclude any chemical effect. On the other hand, the typical saliva reaction could not be produced through quartz; with yeast on one side and saliva on the other, only partial effects could be obtained, such as absence of granulation, or tendency to become spherical. The pictures were never convincing, and it must be left to a later investigation to solve this problem.

This harmful effect is not typical for all saliva. It is typical for the individual, and pratically independent of the diet. An investigation of the saliva reactions of several members of a family showed the above-mentioned injurious effect with male members of the family, and one female member. Two other females stimulated yeast growth, produced elongated forms, and, with *Saccharomyces mycoderma punctisporus*, a manner of growth strikingly resembling mycelium.

C. NECROBIOTIC RAYS

Lepeschkin had observed (1932a) that the stability of living matter (plant cells, erythrocytes) is increased by irradiation with weak ultraviolet light, while strong intensities made cells less stable. The two effects are independent of each other. He concludes (1932b) that weak intensities of ultraviolet must help in the synthesis of cell constituents, and that *vice versa*, when cell constituents break down, the energy absorbed during synthesis must be released again, emitted, partly at least, as ultraviolet radiation.

The experimental proof is given in considerable detail in a later paper (1933). The test organism was nearly always yeast, though parallel experiments were made also with leaves of *Elodea*, with petals of flowers, e. g. of *Papaver*, and with suspensions of *Bacillus subtilis*. When silver nitrate was added to a living yeast suspension in the dark room, the yeast died within 12 minutes, and the suspension was gray. When the yeast had been killed by ether or by heat before the silver salt was added, the suspension remained white, but turned gray upon exposure to light. The gray color of the silverprotein precipitate with living yeast was supposed to be brought about by ultraviolet rays emitted by the dying cells. A similar difference could be observed when yeast was suspended in a mixture of solutions of KBr and $AgNO_3$. Living yeast with ether caused a dark discoloration, but when the yeast had been killed by ether before the AgBr mixture was added, the mixture remained light-colored.

Since these experiments did not exclude chemical effects, AgBr suspensions in small quartz tubes were inserted into tubes with dead yeast, and also into those with living yeast plus ether. After exposure with continuous shaking in the dark room, the AgBr suspensions were removed and mixed with a photographic developer. In all experiments, the suspension exposed to dying cells proved to have received some radiation.

Then, very sensitive photographic plates (Eastman Speedway) cut in small strips, were submerged directly into the yeast suspension. Part of the plate was covered with filter paper which excluded physical, but not chemical effects by soluble substances. After 10 to 25 minutes exposure to yeast previously killed by ether, the plates upon development remained light. With living yeast, they were also light. However, when ether was added to the living yeast, the dying cells affected the plates so that during developing they turned dark, except for the little strip shaded by the filter paper. This proved to LEPESCHKIN's satisfaction that the effect was physical and not chemical.

From the absorption of these rays by glass and by gelatin, LEPESCHKIN estimates their wave length to be largely between 1800 and 2300 Å, with a very weak emission of greater wave lengths. This agrees fairly well with the range of mitogenetic rays. From the above experiments, it seems as if the necrobiotic rays were stronger than those from actively fermenting yeast cells. LEPESCHKIN could obtain an indication of an effect upon AgBr suspensions if he used living beer yeast instead of baker's yeast, and added 10% sugar. However, though there was a slight effect from the fermentation upon the AgBr, the effect from the same mixture with ether i. e. with dying cells was much stronger.

LEPESCHKIN then ventures further to state that many of the mitogenetic phenomena are in reality due to necrobiotic rays. He emphasizes the radiation of necrobiotic processes e. g. autolysis and of wounds as proof for his contention. Evidently, LEPESCHKIN was not familiar with the latest literature on this subject. In the case of wounds, it is not the injured cells which radiate, but the uninjured cells next to the wound. The radiation spectra of the various chemical processes are ample proof that dying cells are not necessary for the production of mitogenetic rays. LEPESCHKIN apparently feels this; he believes it possible that "necrobiotic rays" might also be emitted from a decomposition of vital compounds in living cells which might be imagineable during very rapid physiological processes. He mentions respiration as a possibility. However, it would be necessary to consider almost all exothermic reactions as necrobiotic processes in order to combine the two types of radiation.

SUCHOW and SUCHOWA (1934) have perhaps found the link between LEPESCHKIN's and GURWITSCH's explanation. They conceived the idea that the "necrobiotic rays" were emitted from the coagulation of proteins, and they tested it by coagulating egg white by alcohol in quartz or glass vessels which were placed over AgBr-suspensions as in LEPESCHKIN's experiments. The experiment was carried out in the dark, and after exposure, the two suspensions were brought into light. In 25 experiments, the suspension which stood under the quartz vessel uniformly darkened sooner than the other.

D. INFRA-RED RADIATION

It might appear rather probable that some organisms would emit infra-red rays since some are capable of producing visible and ultraviolet rays. The only case of near infra-red emanation known to the authors is, however, a series of observations by STEMPELL (1931) that sprouting peas will increase distinctly the rate of spontaneous decomposition of a saturated solution of H_2O_2. The effect passed through glass, and was not visible, it must therefore be of an infra-red nature. The temperature of the peas rose 0.5^0 by their own respiration, but an artificial increase of 5.5^0 was necessary to bring the rate of peroxide decomposition to that produced by peas.

Equally rare are observations of an effect of infra-red rays upon living organisms. The only one known to the authors is an experiment by NELSON and BROOKS (1933). They exposed the unfertilized eggs of 2 sea urchin species and of one worm to infra-red rays of 8000 to 12 000 Å, obtained from a Mazda lamp by means of a monochromator. After 15 to 45 minutes exposure, the eggs were fertilized by the usual method. The irradiated eggs showed, in each of the 9 experiments a distinct decrease in the percentage fertilization. The decrease varied between 18.5% and 87.1%. The temperature difference between control and exposed eggs was not more than 0.06^0 C. The authors believe therefore that the reduced fertilization is caused by a photochemical effect.

E. BETA-RADIATION

An entirely different type of radiation should be mentioned only in passing, namely the beta-radiation of potassium. Though it has been proved experimentally, and is of importance to life processes, it will not be discussed extensively here because this type of radiation is utterly unlike the mitogenetic and related rays. It originates from the radioactive fraction of potassium, and is really not characteristic of the living cell, but of its potassium content. The intensity of this radiation is the same whether the cell is alive or dead.

Nevertheless, this radiation is biologically important, and may be the chief reason for the indispensibility of potassium in living organisms. ZWAARDEMAKER (1921) was the first to test experimentally the physiological importance of potassium radiation. He succeeded (1926) in keeping isolated frog hearts beating by substituting the potassium of RINGER's solution by radioactive equivalents, rubidium, uranium, thorium, radium or ionium. In 34 experiments, he could show that frog hearts which had ceased to beat in RINGER's solution minus potassium, started again in the same solution after about half an hour's irradiation by mesothorium (in glass) or radium (through mica). After removal of the radioactive substance, heart beat soon ceased again and could frequently be brought back a third time by new exposure to beta rays.

SCOTT (1931) determined the total energy from potassium of the average human heart to be 9.64×10^{-6} ergs per second. He states that "one may readily conceive that the free energy of the beta particles can be cumulative and, reaching a maximum, transform the potential energy of the heart muscle in response to node and bundle impulses into the enormously greater manifestation of kinetic energy, the systolic contraction".

CHAPTER V

SPECIAL PROPERTIES OF MITOGENETIC RADIATION

The preceding chapter has revealed that the socalled mitogenetic radiation is a real radiation according to the strict physical definition of the word. It travels through space rectilinearly; it can be reflected (pp. **59** and **80**); it can be absorbed (p. **59**); it shows refraction and dispersion (p. **35**).

The present chapter describes some peculiar properties of mitogenetic rays which are not common to all types of radiations.

A. INTERMITTENT IRRADIATION

Very early in the history of mitogenetic radiation, it was discovered that the effect could be intensified by irradiating intermittently instead of continuously. The customary method for this purpose is the insertion, between sender and detector, of a rotating disk which contains one or several openings or windows. The width of these, their number, and the rate of rotation of the disk allow one to vary the three items concerned in intermittent processes, i. e., the frequency of exposures, the duration of each, and the total time of actual irradiation.

The most striking result with intermittent irradiation is the much shorter total time of exposure necessary to bring about distinct mitogenetic effects. GURWITSCH (1932) determined the threshold value in mutual yeast irradiation by means of disks rotating at approximately 3000 revolutions per minute. The disks contained one or several windows of varying width. The width of the window is measured by the central angle (fig. **39**). From this angle and from the number of revolutions, the duration of each interval and the frequency of interruption can be calculated. Table **31** gives the results obtained. It indicates that the minimal

Table 31

Influence of Frequency of Interruptions and of Duration of Individual Exposures upon the Threshold Value of the Detector Yeast as Sender and Detector (Baron Method)

With continuous irradiation, 6—8 minutes exposure was required for a mitogenetic effect

The disk contained	Duration of individual exposure, in sec.	Frequency of exposures per sec.	Total number of exposures	Total time of actual exposure in sec.	Mitogenetic effects
16 windows of the angle 2.5^0 . .	0.00014	800	70 000	10	5; 1; —1
16 ,, ,, ,, ,, 2.5^0 . .	0.00014	800	91 000	13	50
16[1] ,, ,, ,, ,, 2.5^0 . .	0.00028[1]	400[1]	23 000	6.5	10; 8
16[1] ,, ,, ,, ,, 2.5^0 . .	0.00028[1]	400[1]	46 000	13	37
8 ,, ,, ,, ,, 10.0^0 . .	0.00050	400	25 000	12.5	40; 51
2 ,, ,, ,, ,, 20.0^0 . .	0.00100	100	13 000	13	22; 23; 27
2 ,, ,, ,, ,, 20.0^0 . .	0.00100	100	20 000	20	50
1 ,, ,, ,, ,, 7.5^0 . .	0.00050	50	30 000	15	—1; 2
1 ,, ,, ,, ,, 10.0^0 . .	0.00070	50	19 000	13	42
1 ,, ,, ,, ,, 10.0^0 . .	0.00070	50	23 000	16	1; 2
1 ,, ,, ,, ,, 10.0^0 . .	0.00070	50	40 000	30	27; 28; 29; 26

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[1] The disks in these experiments were running only half speed.

actual exposure must be more than 10 seconds, 12.5 to 13 seconds being sufficient. When the same experiment was tried with uninterrupted irradiation, it required 6 to 8 minutes to produce a distinct effect. The rhythmic interruption of radiation had decreased the threshold time to about 1/30th of the amount required with continuous exposure.

The frequency of interruption has some bearing upon the threshold value. When the frequency is between 800 and 100 per second, 13 seconds are sufficient for induction. However, when it falls to 50 per second, 15 and 16 seconds of total exposure are usually insufficient, and 30 seconds are necessary to show a mitogenetic effect. This is to be expected, since it signifies an approach to continuous irradiation. Zoglina (quoted from Gurwitsch 1932, p. 256) repeated the same experiments with a half-disk rotating at 10 r.p.m. This meant uniform intervals of exposure and irradiation of $^1/_{20}$th second each. The following percentages in increase of buds were obtained with a total exposure time of

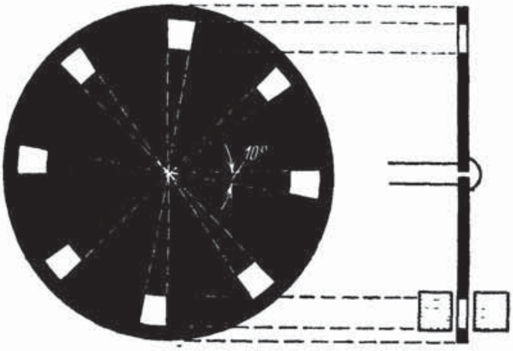

Figure 39.
Rotating disk for intermittent radiation. at right: side view showing the position of the two agar blocks with the yeast sides facing each other, for intermittent muto-induction.

50 seconds:	— 7	—15	0	— 5	+ 4
60 „	+28	+56	+64	+28	+36

The increase in efficiency of a photochemical reaction by intermittent irradiation has its analogy in the increase of photosynthesis of green plants by intermittent exposure. Warburg (1919) measured the amount of CO_2 absorbed by an alga, Chlorella, during 15 minutes of actual exposure, either continuously or discontinuously. A rotating disk was used which was in principle like that of fig. 39, but the times for light and dark were made equal. The results are given in Table 32. An increase up to

Table 32

CO₂-assimilation by an alga, with continuous or intermittent exposure to light

(exposure was either 15 minutes continuously or 30 minutes discontinuously, with alternating light and dark intervals of equal lenght)

Number of revolutions of disk per minute	Duration of each individual exposure	Total Number of exposures	Strong Light		Weak Light	
			CO_2 absorbed in mm³	Increase by intermittent light %	CO_2 absorbed in mm³	Increase by intermittent light %
0	15 minutes	1	31	—		
2	15 seconds	60	37	14		
0	15 minutes	1	34	—		
20	1.5 seconds	600	45	36		
0	15 minutes	1	32	—		
200	0.15 seconds	6 000	50	56		
2000	0.015 ,,	60 000	55	72		
0	15 minutes	1	46; 49	—	45: 45	—
20	0.375 seconds	2 400	70	46	—	—
200	0.0375 ,,	24 000	85	77	—	—
2000	0.00375 ,,	240 000	94	96	42; 43	0

practically the double amount was observed with strong light, but no difference with weak intensities[1]).

WARBURG considers two possible explanations: either assimilation continues for some time after darkening (e. g. through some short storage of energy); or, assimilation is more rapid at the first moments of exposure because more substances have accumulated ready for photosynthesis while later, their concentration is only comparatively small. His intention to investigate in more detail the latter, more probable case seems never to have materialised.

It is not at all certain that this observation is really analogous to the increase of the mitogenetic effect. WARBURG could double the amount of photosynthesis by distributing the same total radiant energy over twice as long a period. GURWITSCH, however, found a 30-fold increase in the threshold value while the greatest difference between light and dark periods was only 1 : 9. Besides, there seems to be no difference between strong and weak intensities. On the other hand, we cannot be certain that these threshold times are reliable measures of intensity of radiation (see p. **114**).

The greatly intensified susceptibility of the living detectors by rhythmic discontinuity of radiation shows that by this method, radiations can be detected which otherwise produce no effect whatever when applied continuously. This holds true not only with very weak senders, but also with very strong sources which produce either no effects or depressions (see p. **115**). The greater susceptibility also permits transmission over longer distances. While mutual induction of yeast has its limits at 3—4 cm. with continuous exposure, very good results can be obtained over 15 cm. with intermittent irradiation (GURWITSCH 1932, p. 260). These facts differ greatly from WARBURG's observations with algae, where weak intensities of radiation could not be induced to produce stronger photosynthesis by intermittent exposure (Table 32).

A *rhythmical* interruption of the radiation seems to be essential for mitogenetic induction. Parallel experiments were made with two disks rotating at the same speed, both with a total window width of 75°. In one disk, the 75° were distributed uniformly; the other disk contained openings, varying in size from 2.5° to 30°, in irregular distribution. Only with the regular spacing were positive results obtained (GURWITSCH 1932, p. 261). This, could hardly be accounted for by any of the two explanations of WARBURG's.

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[1]) After the manuscript was finished, a paper by EMERSON and ARNOLD (1932) came to our notice. They were able to increase photosynthesis 400% by intermittent irradiation whereas WARBURG (1919) could only double it.

B. INFLUENCE OF DIFFUSED DAYLIGHT

Many of the common "senders" of mitogenetic radiation affect other organisms only when in daylight. Diffused light is entirely sufficient. This has been observed for onion roots cut off from the onion bulb, and for the pulp of the onion base, as well as for the pulp of a number of plant tissues. In 1930, POTOZKY gave several series of experiments showing that the same holds true also for yeast. The experiments were made by the Baron technique, measuring the percentage increase of buds on yeast grown on agar blocks. All experiments were made by muto-induction of yeast, i. e. by exposing yeast to yeast.

Table 33. Influence of Daylight upon the Yeast as Sender and as Detector of Mitogenetic Effects Obtained by Muto-Induction

Sender:	Dark Yeast		Daylight Yeast		Dark Yeast		Daylight Yeast	
Detector:	Dark Yeast		Dark Yeast		Daylight Yeast		Daylight Yeast	
exposed in	daylight	dark	daylight	dark	daylight	dark	daylight	dark
Percentage	0.7	16.0	21	—8.0	0	—4.5	35	9
Increase	5.4	3.3	22	—8.6	3.2	—1.8	30	—8
in Buds	5.5	4.3	20	—2.9	3.1	—3.6	30	—9
	—11.0	2.2	23	—3.9	1.0	—1.8	25	1.8
	2.1		30		11.0		24	—15
	1.8		30		9.4		29	6.3
	3.3						20	9
	2.3						22	7
Average . .	+1.4	+6.5	24.0	—5.9	+4.6	—2.9	27.0	+0.2

Three factors were varied: the sender yeast, the detector yeast, and the light during exposure. Yeast grown in the dark had no effect upon yeast, whether grown in the light or the dark, and whether tested in light or dark. Yeast grown in daylight showed distinct radiation and mitogenetic effect upon the detector yeast, regardless of whether this had been grown in light or dark, but only when the exposure was made in daylight. This accounts probably for a number of negative results by some experimentors.

Yeast grown in the dark regained the property of radiation after remaining in daylight for about two hours.

Frank and **Rodionow** have shown, by means of a **Geiger** counter, that light affects greatly radiation from some chemical oxidations, as e. g. $K_2Cr_2O_7 + FeSO_4$ (p. **34**).

C. SECONDARY RADIATION

The original experiments by **Gurwitsch** had shown that only the meristem, i. e. the growing tissue near the tips of onion roots radiated while the older parts of the root, where the cells had ceased to multiply, were inactive. Even the root tips radiated only when connected with the bulb, or at least with part of it. They lost their radiation completely when severed from the bulb.

In search for an explanation, **Gurwitsch** discovered that a root, after being cut from the bulb, will emit a radiation when it is exposed to ultraviolet light. This "secondary" radiation of the root ceases when the "primary" radiation does. It seemed quite impossible that the original rays as such could have been transmitted through the root by reflection without having been absorbed completely. A chemical effect could hardly be passed along so rapidly. There was only one alternative left: the radiation from the outside induced the exposed cells to produce some radiation of their own; these "secondary rays" again induced the neighboring cells to radiate, and so the effect was passed along the root without losing in intensity.

This explanation was proved by many variations of the original experiment. For some time, it was believed to be a phenomenon characteristic of the living cells only, until in 1932, A. and L. **Gurwitsch** found it to occur also in nucleic acid solutions, and **Wolff** and **Ras** (1933, 1934) proved it to be primarily a photochemical phenomenon.

Many illuminating details have been worked out by **Potozky** and **Zoglina** (1928), **Alexander**, **Anna** and **Lydia Gurwitsch** and others. Secondary radiation was observed in muscle, in liver, in nerves, in suspensions of yeast, of bacteria, of protozoa. In all these cases, secondary radiation seemed to be glycolytic. The fact that livers from starving animals which are free from glycogen, did not radiate, supports this view. However, this cannot be generalized because the secondary radiation from nucleic acid is *not* glycolytic.

By this mechanism, primary radiations can be spread and transmitted to distant parts of the plant or animal body. It will be shown later that the tips of onion roots are only secondary senders; the primary rays are produced in the onion bulb, by oxidation.

This spreading of the mitogenetic effect can be plainly shown with densely grown agar surface cultures of yeast. Gurwitsch irradiated such a culture through a slit 0.1 mm. wide. When the percentage of buds was counted, there was a distinct increase not only in the irradiated region, but as far as 10 mm. distant. The increase in buds was 65% at the irradiated zone, and at a distance of

1 mm.	2 mm.	3 mm.	4 mm.	5 mm.	6 mm.	7 mm.	8 mm.	9 mm.	10 mm., it was
100%	87%	86%	79%	80%	80%	55%	43%	33%	25% respectively

Experiments with larger irradiated surfaces gave the same amount of spreading, 9—12 mm. from the border of the irradiated area.

The ability of roots to produce and conduct secondary radiation is limited to a short time after the severing of the root from the bulb; Potozky and Zoglina (1928) found a positive effect after 30 minutes, but not after 40—45 minutes.

The same authors could also show that the production of secondary radiation exhausted the plant rapidly. Freshly-cut roots which gave strong secondary effects during the first 5 minutes of irradiation showed no reaction after 10 more minutes of exposure to monochromatic light of 2020 Å.

Another experiment with starving yeast cells may help to throw some light on this phenomenon. Yeast cells radiate immediately after being washed, but not 30 minutes later. Even then, the organisms will still produce secondary radiation under the influence of an arc light spectrum. This exhausts the yeast so much, that one hour later, it has produced 41% less buds than the unirradiated control. Exhaustion has also been demonstrated with chemical solutions (see p. 44).

A very recent illustration for such exhaustion has been given by Latmanisowa (1932) on the secondary radiation from nerves after mitogenetic irradiation. The sciatic nerve of a frog was irradiated by a yeast culture. Another yeast block, serving as detector, was placed near the irradiated part of the nerve so that it was exposed only to secondary radiation from the nerve, but not to primary radiation from the yeast. This block was changed every 5 minutes. Fig. 40 A shows that the induction effect of secondary rays is very strong at first, but decreases after 5 minutes, and after approximately 30 minutes, it has disappeared, and does not appear any more upon continued irradiation.

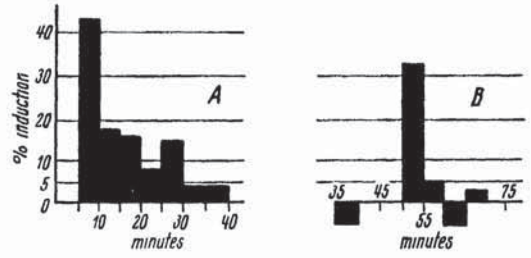

Figure 40.
Secondary radiation from a nerve exposed to continuous yeast irradiation. *A*: the first 40 minutes, showing exhaustion of the nerve; *B*: a nerve, exhausted after 35 minutes, is given 10 minutes rest after which irradiation is continued.

If, however, the nerve is given a "rest" for 10 minutes, by removal of the source of irradiation, it will recover sufficiently to react again upon renewed irradiation (fig. 40 B). But the nerve is still "tired" and will become much more readily exhausted than the first. This phenomenon, too, is not characteristic of nerves only. It can be duplicated with cell-free solutions (p. **43**).

The observation that secondary radiation could be passed on over considerable distances, suggested measuring the rate of travel. After some preliminary experiments by Alexander Gurwitsch, Anna Gurwitsch (1931) made some accurate measurements with onion roots, by means of a rotating disk (fig. 41). This had two windows, one nearer the center through which the primary rays (from a yeast culture) fell upon the older part of the root, and another towards the periphery of the disk through which the radiation from the meristem of the root fell upon the detector. These two slits were so arranged on the rotating disk that after the primary rays had fallen upon the root, the disk had to be turned through 50° before the secondary rays from the meristem could fall upon the detector. This central angle was varied from 20° to 85°. With a definite speed of rotation of 3000 r.p.m., only the angles between 25° and 50° gave positive results. This signifies that a certain time (0.0022 seconds) must pass before primary radiation falling upon one part of the root, is conducted to another part and is emitted there. These values refer to a condition over 2.5 cm. Then, the distance was increased to 5 cm. The central angle for positive effects was hereby increased to between 40° and 70°, which means an average increase of 15°. This corresponds, at 3000 r.p.m., to 0.00083 seconds, and this is the time required for the secondary radiation to pass the additional 2.5 cm. The rate of conduction is therefore about 30 meters per second.

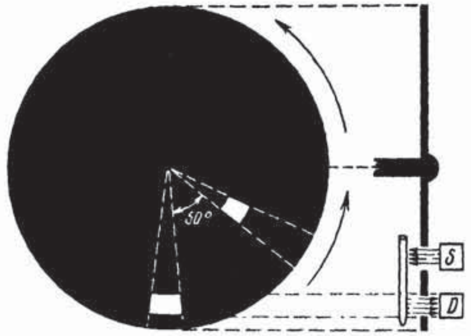

Figure 41.
Rotating disk for measuring the rate of travel of secondary radiation in onion roots. At right, side view showing position of primary sender *S*, onion root, and detector *D*.

Allowing the same time for transmission through the other 2.5 cm. of root, the total time required for transmission through 5 cm. of root is 0.00166 seconds. The total time corresponding to the average angle of 55° is 0.00306 seconds. The difference, 0.00140 seconds, was required for processes other than conduction, such as local reactions at the points of absorption and emission.

Recently, LATMANISOWA (1932) has measured the rate of conduction of secondary radiation in the sciatic nerve of the frog. The method was exactly the same, except that the 2200 Å area of the copper arc was used as primary source. This was sufficiently intense to permit the reduction of the window for primary radiation to 3° and that for secondary radiation to 1°. The accuracy of the method was thus greatly increased, and the rate of conduction in the nerve was found to be 30±3 meters per second. This is in good agreement with physiological measurements on the rate of conduction of nerve impulses.

In the experiments with secondary radiation of onion roots, radiation was observed from the same side of the root which had been exposed to primary radiation. Further experiments showed that the radiation effect was transferred longitudinally with great ease, but that no conduction occurred transversely across the root to the opposite side. In the nerve, however, strong radiation has been obtained from the unexposed side (LATMANISOWA 1932).

An important phenomenon for the explanation of mitogenetic effects is the observation (GURWITSCH 1932, p. 300) that radiating cells or tissues lose this power rather readily when they are themselves exposed to mitogenetic rays. Even young, actively radiating cultures lose their power when exposed to their own wave lengths. Four yeast agar blocks were placed so as to irradiate one another. The first mutual induction was plainly noticeable. After 15 minutes, they were separated and tested as senders; one was tested at once, the others after 15 and 30 minutes. None produced an increase in budding, as the following data show:

Mutual induction during 15 minutes	37%	increase	over	control
After 15 minutes muto-induction, effect upon new detector	2%	„	„	„
After 15 minutes muto-induction and 15 minutes recovery	0%	„	„	„
After 15 minutes muto-induction and 30 minutes recovery	1.8%	„	„	„

Perhaps 30 minutes is too short a time for recovery of yeast, the generation time of which must have been at least one hour under the condition of the experiment.

This phenomenon itself can be at least partially explained by the experiences with secondary radiation (see p. 45).

Practically all these facts had been discovered, before it was found that in certain cell-free solutions, the same effect can be obtained (see p. 44). This does not alter the explanations materially, however. It only means that secondary radiation need not be connected with life processes. It is brought about by some unknown influence of ultraviolet rays which induce certain chemical reactions in cells or complex organic substances.

In their latest publication (1934a), WOLFF and RAS point out that mitogenetic rays become polarized by reflection like common light does, and that apparently, polarized mitogenetic rays have an enormously greater biological effect. When mitogenetic rays fall upon any cell, it seems highly probable that at least part of this radiation will be reflected from the cell walls, and thus will become polarized. It is not possible, at the present moment, to foresee all the consequences of such polarisation.

D. INTENSITY OF THE MITOGENETIC EFFECT

It has already been stated repeatedly that the intensity of the effect is not proportional to the intensity of the radiation. One very simple reason for this is the usual method of recording the results. The "induction effect" as the increase in the exposed yeast over that of the control, expressed in percentages of the latter cannot possibly be used as quantitative measure, as explained on p. 79.

The error of this method of recording results becomes most evident when applied to physical measurements. If the number of impacts induced by biological radiations is expressed in percentage of the stray radiations of the surroundings, it is utterly meaningless from a quantitative viewpoint because this stray radiation (the background radiation) can be greatly altered by shielding the instrument with iron or lead. This does not affect the intensity of the biological radiation at all. The "mitogenetic effect" as used especially by the Russian investigators has no quantitative value whatever. It is not surprising that it was never possible to use it for measurements of intensities.

The customary way of comparing intensities is to compare the minimal time of exposure (threshold time) required to give definite mitogenetic effects. This method has been used repeatedly by the Russian workers, and recently also by WOLFF and RAS. Examples may be found in Table 12 p. 44, Table 46 p. 145, and Table 49 p. 157. However, there are physical reasons to warn against quantitative conclusions from threshold times. It has been pointed out above (p. 24) that, with photographic plates, the reciprocity law (double intensity means half as long exposure) does not hold with very low intensities.

All previous measurements of intensities have become practically meaningless since WOLFF and RAS (1934a) showed that mitogenetic rays may easily become polarized, and that polarized rays have an enormously much stronger biological effect than the ordinary radiations of this type.

E. RETARDATION THROUGH RADIATION

It seems quite probable that an overdose of radiation might produce the opposite effect of mitogenesis, and prevent or retard mitosis. GURWITSCH called this phenomenon mitogenetic depression which, really, is a self-contradictory term; it should be "depressed mitogenesis". Observations of this kind have been recorded rather frequently, but the circumstance that different detectors sometimes give opposite results, warns against hasty conclusions.

As early as 1928, SUSSMANOWITSCH irradiated onion roots biologically for 12 hours and longer, together with control roots exposed only during the last 2.5 to 3 hours. She observed a decrease of mitoses in the exposed side of the root as compared with the opposite, shaded one. This was interpreted as "exhaustion" by too much radiation. Strong physical light produced the same depresssion in a few minutes.

It must be remembered, however, that with roots as detectors, we have no real controls; a difference between the two sides of the root may mean stimulation on one side, or retardation on the other side, or both. In this case of over-exposure, there may have been retardation through over-exposure, or it may mean no effect through over-exposure, and stimulation (through secondary radiation) on the shaded side.

We must therefore turn to other detectors which permit absolute controls, i. e., to unicellular detectors. The yeast bud method appears to be the one by which "depression" is observed most easily. But it is just these retardations by the yeast bud method which are frequently contradicted, in the same experiments, by parallel measurements of the actual cell increase. Table 34 shows SALKIND's experiments (1933) with rat blood radiation. With exposures of 2.5 minutes and longer, the percentage of buds showed a decrease against the controls, the actual number of cells, however, is larger than that of the controls. This can only mean that the yeast bud technique fails to indicate the true growth rate (see p. 69).

Real retardation by biological radiation can be measured only by decrease in the growth rate. The measurement of the actual number of cells permits of only one interpretation; a smaller increase than in the control can only signify a retardation

Table 34. Induction Effects from Intermittent Radiation of Rat Blood, as Measured by the Relative Increase in Yeast Buds, and by the Increase in Total Cells

Length of Exposure	Induction Effect obtained	
	by yeast buds	by yeast cells
7 seconds	3; 3	
15 „	23; 41; 30	1; 7; 12; 13
30 „	17; 20; 28; 30; 35	47; 69
1.5 minutes	5; 9; 13	
2.5 „	—31; —41	46; 50; 57; 69; 74; 123
5 „	—18; —20; —23; —24; —24; —25; —28; —28; —33	22; 22; 40; 43; 109; 120
10 „	—30; —33; —33; —35	73
20 „	—52	

of the growth rate. Such cases are also reported. Wolff and Ras (see p. 77) consider it the normal reaction after continued irradiation. In his analysis of this phenomenon, Salkind (1933) observed that with prolonged irradiation, the depression did not increase. A more detailed investigation revealed a certain periodicity; after stimulation followed depression, but after depression, if radiation continued, again stimulation could be observed, (Table 35). This was the case with physical as well as biological senders. In each instance, as well in the experiment of Table 34, irradiation was applied intermittently. No *definite* periodicity could be found in Salkind's data.

Gurwitsch as well as Wolff and Ras (1933c) have verified this observation of several maxima at widely different exposure times while between these maxima, no mitogenetic effects were obtained.

A striking parallel exists between this effect and that of the photographic plate, as may be seen by the following quotation from Neblette (1930).

"**Reversal by Light:** With a short exposure to light we get a latent image which on development yields a negative. If the exposure is lengthened considerably, the image becomes positive instead of negative when developed, while still further exposure will produce a second negative, and it is probable that the cycle may be repeated indefinitely, although

Table 35. Periodicity of the Mitogenetic Effect Measured by the Increase in Cell Numbers with Yeast

Source of Radiation	Percentage Increase over Control Cultures					
	Line 2350Å Carbon(or Cu) Arc Light		Agar Culture of Yeast		Serum Albumin in gastric juice	
Experiment No.	I	II	I	II	I	II
Exposed for 18 seconds . .	+48	— 8				
2 minutes . .	— 8	—18	+20	+27	+43	
5 ,,		+ 1	+94	+ 6	+85	+80
8 ,,	+ 8	+28	— 6		+ 2	—23
10 ,,			+38	+39	+77	—38
12 ,,	—13	—19		—11	+17	—32
15 ,,	+37	— 2		+37	+ 7	+37
18 ,,	—40	+30			—16	+88
30 ,,						+79
40 ,,						+40

owing to the enormous exposures required, no one has been able to go past the second negative stage. The reactions which result in reversal are still obscure."

Gurwitsch (1932, p. 219) gives some examples where, after too long an exposure, the effect was not at once harmful, but was delayed for a short time, acceleration being noticeable followed by a distinct retardation. He calls this "secondary depression".

F. ADAPTATION TO GRADUAL INCREASES IN INTENSITY

When the intensity of radiation is gradually increased from below the threshold to a value which would produce a strong effect under usual conditions of exposure, no induction takes place. This has been demonstrated most simply in experiments on mutual yeast irradiation (Gurwitsch 1932, p. 263). An experiment was started with two yeast agar blocks mounted 6 cm. apart, on the movable substage of a microscope. This distance is too far to produce a mitogenetic effect. Very slowly, the two blocks were made to approach one another, until after 5 to 8 minutes, they were very close together; they remained in this position for some time. The total irradiation time corresponded to that of another set with the same yeast culture which had been placed in the final position at the start. While this latter set showed increases of 40 to 50% over the controls, the yeast of the equally long exposed, but gradually nearing agar blocks paralleled the controls. The time required for this slow approach must be about 5 to 6 minutes. When it is reduced to 3 minutes, the regular mitogenetic effect is observed.

The same phenomenon was obtained when an elliptical disk was rotated between two yeast agar blocks. This disk was mounted so that in rotation, it gradually exposed the two agar blocks to each other, and gradually shaded them again. This was sufficient to prevent induction.

CHAPTER VI

ANALYSIS OF THE MITOGENETIC EFFECT

At the present time, the mechanism by which short ultraviolet rays affect living cells is not understood. This chapter does not offer one theory, but presents a number of attempts to account for the various phenomena observed.

These rays were not discovered by chance. From a certain rhythm observed in the division of the sperm cells of amphibia, and in plant roots after special treatment, GURWITSCH (1922) predicted a factor which controlled cell division. From the mode of action, he concluded that this factor could not be chemical, but must be of a physical nature, and in 1923, he succeeded in proving it with onion roots.

Mitogenetic radiation was considered at first merely from the cytological viewpoint, as an emanation produced somehow in the very complicated process of cell division. Only after 1928, when SIEBERT proved this radiation to be emitted also from purely chemical oxidations, the physico-chemical viewpoint entered into consideration. The theories which were developed during the "biological stage" of the discovery have never been fitted completely into the physico-chemical facts observed later. Hence, we lack a clear conception of the working mechanism of these rays.

GURWITSCH has always distinguished between the "*Ureffekt*", the primary effect which is the increase in the number of mitoses, and all other effects of secondary importance. All of GURWITSCH's speculations and explanations start with the results obtained with onion roots; these were the first detectors, and since he made a large number of experiments with them, they are to him probably the most familiar of all detectors.

However, they have the great disadvantage of not offering perfect controls. The number of mitoses in different roots even from the same bulb varies greatly. The customary method is to use the shaded, unexposed side of the same root as control; but we cannot be at all certain that irradiation of one side does not influence the cells on the opposite side as well. In fact, REITER and GABOR claim that they are affected.

The authors of this book who have made no experiments with onion roots, but are familiar with yeasts and bacteria, prefer to start with these simple, unicellular forms as the first objects for an attempt to interpret the primary mitogenetic effect. The best method for this purpose is the yeast bud method by TUTHILL and RAHN (p. 68) where all cells are of the same age; no secondary radiation from older cells complicates the analysis, and the percentage of buds is a true measure of the rate of cell division.

Any interpretation of the mitogenetic effect should account at least for the most remarkable facts observed. The following have been selected as the most important:

(1) The necessity of a particular physiological stage of the cell.
(2) The relation between the intensities of radiation and of effect.
(3) The minimal intensity required for an effect.
(4) The harmful effect of over-exposure.

A. THE NECESSITY OF A PARTICULAR PHYSIOLOGICAL STAGE

This necessity will not appear improbable to a cytologist. The cells of growing tissues are morphologically and chemically quite different from the old, resting cells of the same tissue. This holds not only for the larger plants and animals, but also for cultures of yeasts and bacteria (see e. g. HENRICI, 1928).

When any cell changes from the stage of active cell division to the resting stage, this is caused by some external or internal factors. These factors must be removed, or changed, before old cells can divide again. With unicellular organisms, rejuvenation is brought about by transferring the old cells to a fresh medium. The old cells need from one to several hours before they are "rejuvenated", i. e. able to multiply at the normal rate. This period of adjustment is called the lag phase (see p. 67). In multicellular organisms, old cells can be induced to cell division by wounding, or by unknown outside stimuli as in the case of gall formation in plants, or neoplasma in animals.

It would not appear probable that a resting cell can be induced to a new cell division by a short, weak irradiation. Though we do not really understand physico-chemically the ageing process of a cell, it does not seem likely that the factors which induce ageing could be removed by irradiation. This is borne out by experiment. Mitogenetic effects are not, as a rule, observed with old cells left in an old environment.

The stage of active cell division, does not appear very favorable either for mitogenetic effects. WOLFF and RAS (1932) make the unrestricted statement that mitogenetic effects can be obtained only during the lag phase, but not later, i. e. not during the phase of constant growth rate. Their experiments support this claim. They explain it by the assumption that the rapidly multiplying cells irradiate one another, and being so close together, their own radiation is stronger than that from any external source, which is necessarily weakened by distance and absorption.

If this explanation were correct, such cultures should react to outside irradiation at low temperatures where the rate of metabolism, and consequently the intensity of radiation, is weak; they should also react to an external source when they are widely dispersed so that the cells are far apart. The latter was tried without success by FERGUSON and RAHN (1933). Cultures of *Bacterium coli,* 24 hours old, never reacted upon irradiation, whether exposed as such or diluted 1 : 10000, while older cultures gave very pronounced effects. The fact that actively dividing cells do not respond readily to mitogenetic rays is thus verified, but the explanation by WOLFF and RAS is doubtful.

With yeasts as well as with bacteria, the stage of rejuvenation, the lag phase, is one of strong response. Very striking are the results of TUTHILL and RAHN (Table **21**, p. **68**) where the yeast produced buds very rapidly when exposed within half an hour after being transferred to the fresh nutrient medium, but failed to respond an hour later, though the control had not as yet started to produce buds. There seems to be one stage during rejuvenation when the cells are most susceptible.

The explanation may be cytological, chemical or physical. It may be that but one mitotic stage can take advantage of the energy introduced into the cell by this radiation. Perhaps, a certain chemical process in the rejuvenating cell is greatly stimulated; e. g. the ultraviolet, by means of a chain reaction, might set up the reduction potential necessary for normal cell functions (light the candle which then keeps on burning as long as the cell feeds normally). Or, possibly, the cell wall becomes transparent to these rays only at one certain stage of development. Whatever be the explanation, it must be kept in mind that so far, the later stage of active cell division does not seem to be greatly influenced by these rays. It appears that the difference between rejuvenation and active cell division might give us the clue for the mitogenetic effect.

There seems to be another stage where cells respond, namely immediately before entering the resting stage. The description of the physiological condition of BARON's yeast plate (p. 66) suggests this strongly. The volumetric method as described by KALENDAROFF (p. 73) appears to make use of this stage, and so does HEINEMANN's hemacytometer method (p. 72). Apparently, the cells, at the point of going to rest, are stimulated to at least one more cell division by irradiation. This may also be the cause of the mitogenetic effect in onion roots (see p. 129). This need not necessarily involve a mechanism different from that assumed in the rejuvenation process. It may well be that in the ageing cell, the additional, properly dosed energy from the sender prevents a certain phase of the ageing process, for a short time, sufficiently long to permit one more cell division. This may be the same mechanism which, under the more favorable conditions of rejuvenation, is stimulated so greatly.

B. RELATION BETWEEN THE INTENSITIES OF RADIATION AND OF EFFECT

It has been one of the most annoying puzzles of mitogenetic experiments that there seemed to be no proportionality between cause and effect even when polarisation is excluded. We could not expect this with detectors involving secondary radiation by old cells, such as the onion root, BARON's yeast plate, or the volumetric yeast method. But even with the yeast plate of TUTHILL and RAHN, where mutual cell influences are practically excluded, the percentage of buds was not at all proportional to the length of irradiation time. When freshly prepared detector plates were exposed for different lengths of time, the following percentages of buds were found (after 2 hours of incubation):

	minutes			
exposed for . .	0	10	20	40
	%	%	%	%
through quartz .	21.0	25	36.5	24.5
direct	22.5	23	37.5	27.5

The sender was a 6 hours old yeast surface culture. The exposure of 20 minutes produced a strong effect, either directly or through quartz, 15% more than the control. If there were proportionality, the 10-minute exposure should have produced an increase of approximately 7%, and the 40-minute exposure a 30% increase. Neither of these other times showed any great effect, however.

This may be explained by the recent discovery of WOLFF and RAS (p. **43**) that nutrient media produce secondary radiation when they have been in contact with microorganisms. The entire detector plate begins to emit radiation as soon as it is exposed to a sender. The intensity of secondary radiation does not depend so much upon that of the primary source as upon the chemical composition of the medium. All hope for proportionality must be given up in this case. Only with a medium which does not produce secondary radiation, does a biological measurement of intensity seem at all possible.

C. THE MINIMAL INTENSITY REQUIRED FOR AN EFFECT

All measurements of the intensity of mitogenetic rays are very inaccurate, but the order of magnitude of the strongest senders appears to be about 100 to 1000 quanta per cm² per second. The detector plate by BARON is completely covered with cells, but that of TUTHILL and RAHN has single cells. The probability that a yeast cell of $6 \times 7\ \mu$ is hit in one second, assuming 1000 quanta/cm²/sec, is

$$\begin{aligned} P &= 0.00000042 \times 1000 \\ &= 0.00042 \end{aligned}$$

The probability of being hit in one minute is 0.0252. It will require 40 minutes of continuous, uniformly dispersed radiation before each of the yeast cells is likely to be hit by one quantum of ultraviolet light. Since we find the strongest effect under this arrangement at 20 minutes (see above), it would seem that one quantum per cell is sufficient to produce the mitogenetic effect. There has been a good deal of speculation as to the mechanism by which one single quantum could affect the cell so greatly.

However, since certain solutions such as blood serum, or broth in which bacteria have lived or are living, will produce secondary radiation, the assumption of a single quantum acting upon the cell has become unnecessary, even improbable. The raisin agar upon which the yeast is spread will gradually become transformed into a secondary sender by the very presence of the yeast. Irradiation will then set the entire mass of agar radiating, and the number of quanta thus produced, or absorbed by the cells, cannot be estimated.

It is known that yeast cells, or onion root cells, or pulp of tissues, yield secondary radiation; it seems safe to assume that living protoplasm generally will respond in this way. Then, if the cell absorbs one or a number of quanta of ultraviolet, the entire cell begins to radiate, not visibly, but measurably. This induces a state of excitement, and it is quite probable that a more rapid cell division may be brought about, provided that the cell is at the proper cytological stage. Possibly, the synthetic powers of the cell work to a certain morphological and physiological culmination which can be released only by a very accurately measured impulse, i. e. the absorption of one quantum of ultraviolet of fairly definite wave length. Considering the systematic arrangement of all molecules in the cell, it can be well imagined that such a release will start many wheels turning, many processes going on automatically and exothermically, until cell division is completed.

This is, in slightly different terms, GURWITSCH's original conception of the mechanism of the mitogenetic effect. He claimed, and seems to assume even now that no cell division is possible without this external, ultraviolet stimulus. It would appear that single-cell cultures of bacteria and yeasts were a proof against this assumption, but they may be explained in some other way.

One fact, however, makes the above explanation too simple. Every fermenting yeast cell liberates, within the cell, energy of definite mitogenetic wave lengths, namely of 1900, 1910, 1930, 1950 and 2170 Å (p. **37**). If all yeast cells produce this wave length, how can cells be stimulated by the same wave lengths from an external source? We may return to the first of our fundamental facts that only at a certain cytological stage, cells will react to mitogenetic radiation. No reaction has been observed at the stage of most active multiplication which is also that of most active metabolism. A very definite and strong response was obtained at the first stage of the rejuvenation process. If we could make the assumption that during the period of sensitivity, the cells show no metabolism, or at least emit no ultraviolet, then the mitogenetic effect could be easily explained. But the assumption is not justified. RAHN (1928) and RAHN and BARNES (1932) found that old yeast cells, compressed baker's yeast as well as beer yeast stored for several weeks at low temperature, fermented strongly within 10 minutes after being placed in sugar solution.

Whatever the explanation, it is certain that the mitogenetic effect does not occur merely through the increase in energy content of the cell.

D. THE HARMFUL EFFECT OF OVER-EXPOSURE

The harmful effect of over-exposure is more easily understood by the secondary radiation of the cell and of the medium. Before this was found, the stimulating effect which the first quantum had produced, appeared to be counteracted by the absorption of a second quantum. Now we realize that the first as well as the second quantum are probably multiplied manyfold within and outside the cell.

It has been shown (p. **43**) that too long exposure destroys the ability of a solution to produce secondary radiation. After a day or two of rest, this property returns. Nothing is known about the chemistry involved, but the assumption of an equilibrium, disturbed by irradiation and slowly re-established after discontinuance, fits best into our present conceptions of life functions.

Something similar to these effects may happen in the cell. We have already seen that very likely they are all capable of secondary radiation. They also will become exhausted upon long-continued exposure (p. **110**). Moreover, it has been shown that recovery is slow. If we assume that all cells are brought to a state of radiation, or excitation, but that only those cells which are at the proper cytological stage can respond to this stimulus by dividing more rapidly, the cells of other stages will soon become exhausted. This would mean at first a normal rate of cell division, and eventually a temporary interruption of mitosis, on account of exhaustion of certain chemicals in the cells, by the prolonged secondary radiation.

Since exhaustion of solutions lasts for a day or two (p. **43**) and exhaustion of cells for hours (p. **110**), it would be difficult to explain the periodical alternation of stimulation and depression observed with long-continued irradiation by Salkind (p. **116**). The data of Wolff and Ras (Table 25, p. **77**) also seem to indicate recovery from depression though irradiation is continued.

The time during which mitogenetic effects can be observed seems to vary with the detector. The sharpest limitations observed are those by Wolff and Ras (Table 12, p. **44**): strong positive effect with 5 minutes exposure, none whatever with 4, 6, 7 or 8 minutes, etc. This is doubtless caused by the uniform age of all cells in this kind of detector while Baron's yeast plate with cells of many different physiological stages, has a more gradual range of response and tolerance.

E. STORAGE OF MITOGENETIC CHARGES

Ferguson and Rahn (1933) observed that bacterial cells could be kept in their old environment for 2 hours after exposure to mitogenetic rays, and still show stimulation of growth when transferred to a fresh medium (Table 36).

Table 36. 3-day old culture of *Bacterium coli*, irradiated by an agar culture of *Bacterium coli* for 30 minutes

	transplanted immediately after irradiation		transplanted 2 hours after irradiation	
	Control	Exposed	Control	Exposed
start	4 950	5 050	4 350	3 950
after 2 hours .	4 950	5 750	3 700	5 900
„ 3 „ .	5 100	6 500	—	—
„ 4 „ .	5 500	8 700	3 650	5 200
„ 6 „ .	24 500	83 500	10 500	44 000
„ 8 „ .	234 200	1 500 000	—	—

The storage of energy as such appears out of the question. A continued internal secondary radiation is also impossible. We can only assume that the cells were changed chemically, that the unknown process of rejuvenation was released, but could not materialize under unfavorable environmental conditions; as soon as this situation was altered, rejuvenation set in at once. This observation may eventually help to locate the exact process released by the mitogenetic impact.

F. MECHANISM OF THE BARON YEAST DETECTOR

In his monograph, GURWITSCH devotes 50 pages to the analysis of mitogenetic effects in the BARON yeast plates. The limited space of this book does not permit detailed quotation, especially since the complexity of this detector leaves too many possible explanations. However, since it is a good parallel to multicellular detectors, we shall at least give a brief summary here.

The complex structure of the BARON yeast plate, with old cells, beyond the stage of cell division, on the surface, and with normally-dividing cells at the bottom, has been described in detail on p. 66. The old surface cells react upon irradiation by producing secondary radiation. They can emit only a definite (though unknown) amount of radiation. After that, they are exhausted and remain inactive, though absorbing ultraviolet, for the duration of the experiment. The number of these secondary senders decreases therefore gradually during exposure.

The absorption of one quantum is sufficient to induce secondary radiation in a surface cell provided that the cell is not too old. Secondary radiation consists in the emission of a number of quanta. Since the emission takes place in all directions, the intensity decreases very rapidly with the distance from this cell. However, since in this detector, cells are lying closely side by side, a quantum emitted through secondary radiation may be absorbed by another reactive cell which then, on its part, emits new secondary quanta. On p. 110, it has been shown that in these yeast plates, mitogenetic effects may be observed 10 mm. removed from the exposed cells. Some of these secondary quanta will penetrate into younger cells, and stimulate them to bud formation.

Gurwitsch considers the "mitogenetic field" similar to an electro-magnetic field. He believes that a uniform stream of quanta striking a cell from all sides will not produce a mitogenetic effect. The mitogenetic stimulus consists in the one-sided discharge (release) of a neighboring secondary sender. Gurwitsch assumes that if two or more quanta strike the surface of the detector at the same moment, the peripheries of the streams of secondary quanta may be partly superposed and thus by interference, produce no effect; though the total energy is increased, the potential necessary to initiate cell division is lacking. Such "equalization" will occur more commonly with physical sources of ultraviolet because the light is more uniform, while in biological sources, radiation comes from many cells unevenly distributed. Consequently, there is less equalization, and therefore a relatively stronger mitogenetic effect must be expected from biological senders.

By irradiating a liquid bacterial culture in quartz from above and below at the same time, Miss Ferguson, in an unpublished experiment, obtained a strong mitogenetic effect. The two radiations did not cancel. Such interference seems rather improbable if we look at the mitogenetic effect as a photochemical one. We would not expect the reaction between hydrogen and chlorine (p. 46) to be suspended if the gas mixture were irradiated from two opposite sides. Such interference is imagineable in a one-dimensional system, e. g. a nerve fiber, but hardly in three-dimensional media.

G. MITOGENETIC EFFECTS IN MULTICELLULAR ORGANISMS

There is one essential difference between the cells of unicellular and multicellular detectors which must be kept in mind to prevent misleading generalizations. Bacteria and yeasts in the detectors mentioned have a very large amount of food at their immediate command, whereas in tissues, e. g. in onion roots or in the cornea, the food supply is limited. Thus, in the latter case, there may not be enough food readily available to permit rapid cell division, even after adequate stimulation; or after a premature mitosis, the food may be insufficient for continuing at a subsequent normal rate of cell division.

The one multicellular detector that has been studied cytologically is the onion root. The interpretations of the onion root effect by **Gurwitsch** (p. 55) and by **Reiter** and **Gabor** do not agree. The latter have based their intrepretation upon a cytological analysis of the onion root which deserves attention because it suggests a close analogy of the root with the **Baron** yeast plate. The root can be divided into transverse cross sections which may be designated by the number of successive cells from each section to the tip. This number is fairly uniform whether the successive cells are counted in the center of the root or along the sides. The most sensitive region in respect to mitogenetic rays is approximately 50 to 70 cells from the tip.

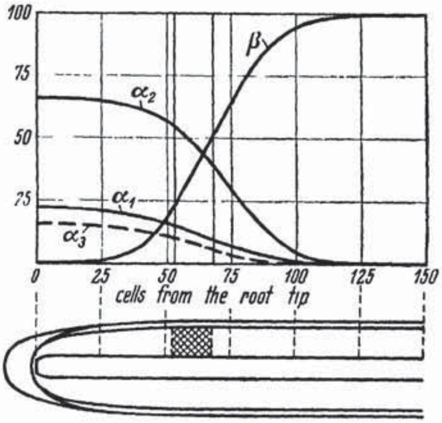

Figure 42.
Distribution of various cells in the onion root.
Below: cross section through the root, with its vascular bundle; the shaded zone indicates the sensitive part of the root.
Above: distribution for the four cell types in different parts of the root; the abcissa represents the distance from the tip, measured by the number of cells.
β = elongated, resting cells; α = short dividing cells. α_1 = newly-born cells, α_2 = ripe nuclei, α_3 = mitotic stages.

Reiter and **Gabor** studied the distribution of cell types along the root. They distinguished the α-type, short actively-growing cells, and the β-type, elongated, resting cells. The first type could be subdivided into 3 nuclear stages: α_1 = newly-born nuclei; α_2 = ripe nuclei; α_3 = various mitotic stages. The distribution of these types is shown in Table 37 and fig. 42. In the root tip, as far as about 25 cell layers upwards, no cells of the β-type, i. e. no resting cells are found. All cells are in active division. In the region which is 125 or more cells distant from the tip, practically all cells are resting; dividing stages are very rare. The zone of mitogenetic sensitivity contains cells of all types, the resting cells amounting to less than half of the dividing types.

To analyze the effect, the authors raised two questions: How much does the exposed side of the root differ from the

Table 37. Distribution of the Nuclear Stages

	a_1 newly-born nuclei	a_2 ripe nuclei	a_3 dividing nuclei	β resting nuclei
at the root tip	21	64	15	0
50 cells upwards from the tip	16	56	11	17
80 ,, ,, ,, ,, ,,	5	21	3	71

opposite, shaded side? and How much does the exposed side differ from the normal? The latter question could be answered only by analogy.

The conclusion was that "under the influence of mitogenetic irradiation, all cells born during the experiment remain in the actively-dividing stage, and produce again ripe nuclei while normally, without irradiation, a certain percentage would go into the resting stage. On the opposite side of the root, more cells go into the resting stage than would normally do so".

Gurwitsch (1932) does not agree with this interpretation. He doubts the possibility of accurately distinguishing between nuclei of resting and dividing cells. He criticizes the method of counting "ripe nuclei" only, and points out that a decrease of mitoses in the opposite side of the root has been observed occasionally, but only in about half of all his (Gurwitsch's) and also of Rossmann's experiments, and this can be accounted for by the method of sectioning and counting.

There is a certain similarity between the onion root thus described, and the Baron yeast plate. Both consist of many closely-packed cells; both have the oldest, non-dividing cells on top, and very young, dividing ones at the bottom. It will be seen later that the onion root is irradiated continuously from above, by the bulb, and this stimulus is transmitted by secondary radiation of the old cells to the young, growing cells in the root tip.

CHAPTER VII

THE SIGNIFICANCE OF BIOLOGICAL RADIATIONS IN BIOLOGY, MEDICINE AND AGRICULTURE

A. UNICELLULAR ORGANISMS

(1) Emission at Different Physiological Stages: It had been emphasized throughout this book that intense mitogenetic radiation has been observed almost exclusively in young, actively growing tissues or cell cultures, while fullgrown ones do not radiate at all, or only rather weakly.

According to GURWITSCH, we must distinguish between two sources of radiation, the one emitted at the moment of nuclear division, and the other resulting from general cell metabolism, such as oxidation, proteolysis, glycolysis. A culture of yeasts or bacteria should be a good sender as long as either the cell division is rapid, or the metabolism remains active. Under optimal conditions of food and temperature, both these functions should cease almost entirely within 24 hours after transfer. Fig. 43 shows the development of a culture of *Streptococcus lactis* at 21°C (RAHN, 1932, p. 401). The left-hand curves represent the multiplication of the bacteria and the gradual accumulation of lactic acid from sugar, by glycolysis. The right-hand curves show the *increases* in each 3-hour interval. These increases must be proportional to the intensity of radiation of the culture, as it is apparent that there can be no other important source of radiation. The bacteria starting with 38 000 cells per cc., can have emitted a noticeable degree of mitogenetic radiation only between 12 and 24 hours. Before that time, the radiation per cell might have been quite as strong or stronger, but the number of cells was too small; while afterwards the cells, though more than a billion per cc., have ceased to produce acid, and therefore to radiate.

With a heavier inoculation, e. g. by flooding the surface of an agar plate with a suspension of yeasts or bacteria, radiation begins sooner because the "active mass", i. e. the number of cells, is greater. At lower temperatures, the intensity is less, but the phase of active radiation is prolonged. With processes which do not result in an inhibiting product like acid, the period of active radiation may also be longer.

This *a priori* deduction is made doubtful by the secondary radiation of the medium in which the bacteria grow. The actual radiation of the bacteria themselves must follow the lines of fig. 43; the emission by the entire culture may not. Intensity of secondary radiation may not be proportional to primary intensity, and besides, since the nutrient medium is continuously changed by the bacteria, its property as secondary sender may change.

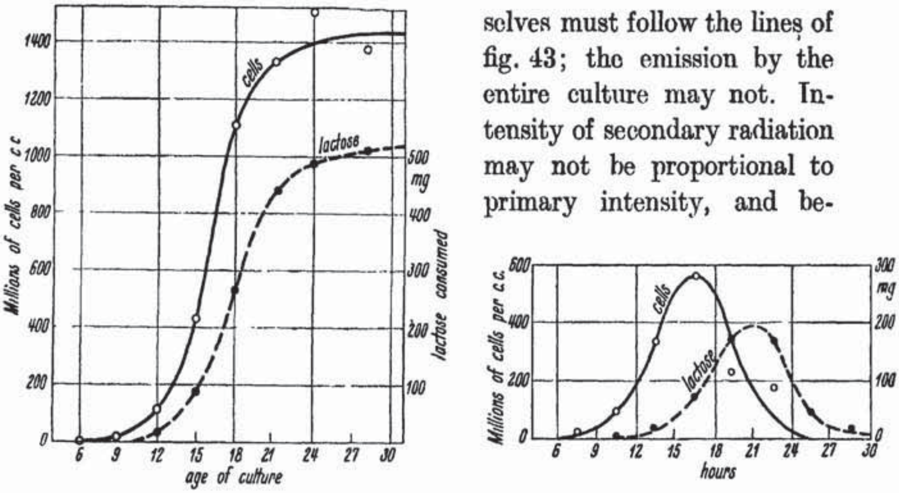

Figure 43. Development of a culture of *Streptococcus lactis* in milk. Age of culture in hours. Full line: number of cells per cc.; dotted line: mg of lactose decomposed in 100 cc.

left: total cells and total lactose fermented; right: cell increase for each successive 3-hour period, and lactose consumed during these 3 hour periods.

However, the theoretical deductions are essentially verified by the observation of Wolff and Ras (1932) that an agar surface culture of *Staphylococcus aureus* remains actively radiating until approximately 18 hours old. When the cultures are full-grown, i. e. when there is no further appreciable increase in cells, nor in metabolic products, the culture ceases to radiate.

Whether metabolism without cell division can produce mitogenetic rays, was decided in the affirmative by a simple experiment of the authors with bakers' yeast suspended in sugar

Table 38. Mitogenetic Effects from Protozoa in Glucose Solution

	Without Sugar	After Addition of Sugar			
		immediately	15 minutes later	20 minutes later	30 mintues later
Opalina	0	—2.6	+40	+36	—
	+4.4	+2.5	+20	+24	—
Paramaecium	+2.7; +2.0	—7	+11	—	— 3.8
	—2.8; +1.0	+7	+12	—	+14.0
	—1.0; +3.0	+3.7	+27	—	—

solution. This yeast cannot grow under the experimental conditions because too many cells are present; however, even if slight growth were to occur, many hours would be required to overcome the lag. During the first hour, therefore, no radiation from cell division is possible while alcoholic fermentation starts at once.

An interesting set of data on the radiation of *Hydra fusca*, the well-known fresh-water polyp, has been given by Blacher and Samarajeff (1930). They found that a pulp of the entire organism radiates. If dissected, the hypostom and the budding zone radiate while the other parts do not. No increase in emission could be detected during regeneration.

The common infusoria do not appear to radiate under normal conditions, but produce a secondary radiation. According to Zoglina (quoted after Gurwitsch 1932, p. 64) the following mitogenetic effects were obtained:

from *Opalina*	0; +4; +3; +6; +12
,, ,, irradiated by blood	+47
,, ,, ,, ,, frog heart	+21
,, ,, ,, ,, spectral light	+15; +19; +38; +50
from *Paramaecium*	+2; +1; +3; —3
,, ,, , irradiated	+20; +22; +13; +40

Radiation could also be obtained from protozoa when glucose was added to the culture; it required about 15 minutes before radiation became noticeable. The mitogenetic effects observed are given in Table 38.

A very interesting claim has been made by Acs (1932). He succeeded in increasing the mitogenetic effect of muto-induction of bacterial cultures (*Bacterium typhi-murium*) by using the detector of one day, i. e. the irradiated culture, as sender for the next experiment. His data show a very consistent increase in the mitogenetic effect through 6 to 7 such transfers.

The proof is not complete, however. It is well known that old bacterial cultures require a longer time to start growing than young ones. The rate of growth of old cultures can be increased for several successive transfers, without any irradiation. Without accurate statements concerning the number of cells in the sender and in the detector, no definite conclusions can be drawn.

(2) Reaction Upon Irradiation: Bacteria and yeasts, when transferred from an old culture to a fresh medium, do not start growing at their maximal growthrate. The old cells undergo a rejuvenation process, morphologically and physiologically (see p. **120**). This results in a very slow growthrate during the first hours after transfer. The bacteriologist calls this the lag phase.

It has been observed by RAHN (1907), PENFOLD (1914) and others that bacteria after being transferred from an old culture rejuvenate more rapidly if many cells have been transferred while with a few cells in a large volume of fresh medium, rejuvenation is slow, and single-cell cultures frequently die. This observation had been very puzzling to biologists since the opposite should be expected, until it could be explained as a simple stimulating effect by mutual irradiation of the cells. The closer they are together, the stronger must be the effect, the quicker the recovery.

The best example is very likely that of HENRICI (1928, Table I) with yeast. He inoculated increasing amounts of yeast into flasks of sugar-peptone solution, so that all the decimal gradations from 2095 to 20 949 750 cells per cc. were present. These different inocula resulted in very different lag periods. The times required to reach the maximal growthrate were:

with an inoculum of	Lag Period	Average cell distance[1])
20 949 750 cells per cc.	2— 8 hrs.	0.036 mm.
2 094 975 ,, ,, ,,	4— 8 ,,	0.086 ,,
209 498 ,, ,, ,,	6—12 ,,	0.192 ,,
20 950 ,, ,, ,,	12—24 ,,	0.422 ,,
2 095 ,, ,, ,,	24—36 ,,	0.914 ,,

---

[1]) The average cell distance is computed from the equation

$$\text{distance} = \text{cell diameter}\left(\sqrt[3]{\frac{74.04}{\text{cell volume in 100 cc.}}}-1\right)$$

It is absolutely correct only with spherical organisms. The volume of one yeast cell is taken as 118 μ^3, the average diameter as 6.4 μ.

Table 39. Growth Acceleration by Mutual Irradiation with Saccharomyces ellipsoideus

Inoculum	Number of Buds per 100 Cells			
	Experiment No. 1		Experiment No. 2	
	8,000 cells	80,000 cells	8,000 cells	80,000 cells
start	0	0	0	0
after 3 hours .	0	0	0	0
after 5 hours .	3	16	—	—
after 6 hours .	—	—	5	49
after 7 hours .	14	60	—	—
after 10 hours .	—	—	17	91
after 12 hours .	—	—	35	91

To these proofs that the lag phase is shortened by the mutual irradiation of cells, BARON (1930) added another. After having shown that bud formation in fresh yeast cultures begins sooner when the inoculum is larger (Table 39), he showed that this difference disappears when gelatin is added (Table 40). Gelatin absorbs mitogenetic rays very completely (see p. 59), and while all other conditions of life remained the same (gelatin contains no nutrients for yeast), muto-induction was prevented, and, as a result, the lag phase became independent of the cell concentration.

Table 40. Prevention of Growth Acceleration by Absorbing the Mitogenetic Rays by Means of Gelatin

Inoculum	Number of Buds per 100 Cells			
	Experiment A		Experiment B	
	8,000 cells	80,000 cells	8,000 cells	80,000 cells
start	1	0	0	0
after 3 hours .	2	2	0	0
after 5 hours .	18	21	12	15
after 7 hours .	30	32	—	—
after 8 hours .	—	—	35	33
after 10 hours .	41	42	48	52
after 12 hours .	—	—	62	61

Table 41

1 cc. of a 3-day old broth culture of *Bacterium coli* irradiated for 30 minutes, then diluted in broth to various dilutions

Dilution	1 : 100		1 : 10 000		1 : 100 000	
Cell Distance	0.140 mm.		0.650 mm.		3.0 mm.	
Cells per	1 cc.		100 cc.		10 000 cc.	
	Control	Exposed	Control	Exposed	Control	Exposed
Start	530 000	460 000	495 000	505 000	570 000	540 000
2 hours	400 000	440 000	495 000	575 000	480 000	490 000
3 hours	575 000	530 000	510 000	655 000	510 000	520 000
4 hours	725 000	750 000	550 000	870 000	540 000	600 000
6 hours	7 850 000	9 950 000	2 450 000	8 350 000	1 570 000	5 130 000
8 hours	29 100 000	23 500 000	23 400 000	150 000 000	17 600 000	70 900 000

Summary of above

Cells per cc. of culture	Average Cell Distance	Cells of Controls in millions		Mitogenetic Effect	
		after 6 hrs.	after 8 hrs.	6 hrs.	8 hrs.
500 000	0.140 mm.	7850	29 100	23	—26
5 000	0.650 mm.	2450	23 400	247	541
50	3.00 mm	1570	17 600	227	303

Table 42. Offspring in 44 hours of the protozoon *Enchelys farcinem* in hay infusion drops of different sizes

Culture A			Culture B		
Drop Weight mg.	Cells after 44 hrs.	Cell Number × Drop Weight	Drop Weight mg.	Cells after 44 hrs.	Cell Number × Drop Weight
5.0	250	1250	1.4	64	90
10.0	31	310	7.4	4	30
13.6	104	1414	13.0	8	104
23.4	64	1498	23.8	6	143
34.6	33	1142	32.2	4	129

The following experiment by FERGUSON and RAHN (1933) verifies this from a different angle. 1 cc. of a culture of *Bacterium coli* was irradiated for 30 minutes, and then diluted in broth 100, 10000 and 1000000 times. The cells in all three cultures came from the same 1 cc., the only difference being their distances from one another after dilution. Table 41 presents the data comparably, i. e. it shows the development of the progeny arising from $^1/_{100}$ cc. of the old culture, when grown in different cell concentrations. The slower growth due to greater dilution is plainly evident in the controls. The more important result is the absence of a mitogenetic effect when the cells are too close together.

(3) Allelocatalysis: The lag phase of growth is not limited to fungi. Quite striking examples of lag in protozoa have been reported by ROBERTSON (1924). This investigator found that single individuals of *Enchelys farcinem* multiplied but very slowly or not at all in small drops of hay infusion while the rate was quite large when two or more individuals were in the same drop.

While this appears a duplication of the lag phase of bacteria, it differs by the fact that the size of the drops has a great influence. Table 42 gives the progeny derived in 44 hours from single cells of *Enchelys* in hay infusion drops of various sizes. In large drops, the growth is much slower, and if extrapolation is permissible, we must conclude that a single cell of culture *A* in 1.5 cc. would not grow at all; with culture *B*, the inhibiting volume is 0.15 cc. This is borne out by experience. ROBERTSON mentions that such cultures usually show no growth.

Robertson assumes that a "catalyst" is produced by the cell, and that a certain concentration of this is necessary to cause cell division. This theory of "allelocatalysis" has not been accepted generally, though similar phenomena are known. **Wildier**'s "bios" (1901) is essentially identical with **Robertson**'s catalyst. The "bios" theory was based, among other things, upon the observation that in the same medium, a small inoculum did not reproduce while a large one did. This was verified by **Naumann** (1919) who observed that 5 yeast cells would die in a medium where 50 produced growth.

It is possible to reconcile the influence of the drop size with mitogenetic radiation. All media absorb mitogenetic rays fairly readily. The ability to grow in a small drop indicates that the cell has been able to produce sufficient radiation to induce mitosis. A considerable part of it must leave the cell, because cells can influence one another mutually. In a small drop, a good share of this emitted radiation is reflected from the air surface and finds its way back to the cell before it is absorbed. With an increase in drop size, distances become greater, absorption is greater and the probability of the rays being reflected back to the cell is smaller. In the isolation of single bacterial cells by the micro-pipette, small drops are an important factor in success (**Wright**, 1929). However, there is no definite experimental proof that allelocatalysis is a radiation phenomenon.

The allelocatalytic effect is not limited to unicellular organisms. A very pretty exemple of mutual stimulation has been observed by **Frank** and **Kurepina** (1930) with the eggs of a sea urchin. With 10 to 20 eggs per drop of sea water, development went much more rapidly than if there were only a very few eggs per drop (see Table 43). It will be shown later that sea urchin eggs at this stage of development are good senders as well as good detectors. However, here too, a chemical mutual influence was not excluded.

B. HIGHER PLANTS

The first mitogenetic sender and the first detector was the onion root. It is established beyond doubt that the tip of the onion root radiates. This radiation is not entirely diffuse, but most of it points quite distinctly in the direction of growth.

Table 43. Development of Sea Urchin Eggs

		Number of Eggs per Drop		
		1—2	3—6	10—20
Experiment I		%	%	%
40 hours after fertilization	not motile	31	6	17
	slightly motile	23	34	9
	distinctly motile	46	40	28
	actively motile	0	20	46
Experiment II				
42 hours after fertilization	not motile	75	0	0
	slightly motile	25	26	4
	distinctly motile	10	40	12
	actively motile	0	34	60
	beginning gastrula	0	0	16
	perfect gastrula	0	0	8

According to GURWITSCH, some negative results obtained with onion roots are very likely due to inaccurate direction of the mitogenetic beam.

Onion roots radiate only as long as they are connected with the bulb, or at least with a part of the bulb. Radiation ceases at once when the root is severed from the bulb. (The roots of the sunflower [*Helianthus*], however, continue to radiate after being cut from the plant; they also continue to grow for a considerable time after being severed.)

This suggests that the substances which are the source of radiation are centralized in the onion bulb, and especially in its base. When the base is cut from the bulb, it radiates weakly. The pulp of the base, however, radiates strongly, and has been used as a strong source of mitogenetic rays in the early experiments. After approximately half an hour, radiation ceases. The spectrum of the pulp is purely oxidative, and its cessation is in all probability due to the completed oxidation of some unknown compound by the oxidases of the root. Heat destroys the radiating power which strongly suggests that an enzyme is the active part (Table p. 38). Heated pulp mixed with exhausted pulp starts to radiate anew, the heated pulp furnishing the compound to be oxidized and the exhausted pulp supplying

Table 44. Radiation Spectra of different parts of the onion

	Induction Effects							
	1900—1950	1950—2000	2000—2050	2050—2130	2130—2200	2200—2260	2260—2330	2330—2410
Onion base pulp	1	—5	+5	—4	+4	+7	+57	+37
Severed roots (secondary radiation) . .	44	43	4	10	37	4		
Intact roots (normal radiation, from tips)	39	32	—2	—2	39	—3	— 3	— 5

the oxidase. Gurwitsch has used the words *mitotin* and *mitotase* for these two essential factors. Considering the oxidative spectrum of the pulp, there can be scarcely any doubt that mitotase is an oxidase. The new terms might better be discontinued because they are likely to be considered as introducing a mysterious new element into life processes.

Since the base of the onion bulb as well as the tip of the root radiates, it seems likely that a relation between these two radiations might exist. Probably, the oxidase is located in the onion base, and the new oxidizable material is transported to it continuously from the leaves through the vascular system of the plant. This accounts for the radiation of the bulb, but not for the emission from the root tips. This question has been decided by spectral analysis (Table 44). The bulb spectrum is oxidative. The normal root tip radiates glycolytically, and cannot therefore be caused by the same process as that of the onion base. The "secondary radiation" of the roots (see p. 110) is also glycolytic, regardless of the wavelength of primary radiation. It seems therefore most probable to assume that the normal radiation of the intact root tip is really a secondary radiation induced by the oxidation processes in the onion base. If this part is narcotized with chloral hydrate, the tips do not radiate. If the base is cut off, the root tips do not radiate. If, however, a small part of the base remains on the root, the tip will radiate.

This conclusion is biologically very important. The conduction of mitogenetic rays through normal tissue over a distance of several inches is bound to affect our conception of growth and of growth stimuli considerably.

Some other evidence supports this explanation. The potato radiates when cut, but only from the leptome fascicles; portions free from leptome are inactive (KISLIAK-STATKEWITSCH, 1927).

A very interesting investigation concerning the radiation of sunflower seedlings has been carried out by FRANK and SALKIND (1926). It was found that radiation can be obtained from the root tips, from the plumulae (the first young leaves) and from the cotyledons. However, only the very center of the cotyledon edge radiated, and no other part of these organs. Failure with one cotyledon which showed an abnormal curvature of the central vein led to the discovery that radiation arises from the vascular system. How mitogenetic radiation is conducted from the vascular system to the growing parts of the plant, the meristem, without great loss of energy, is unknown. It seems improbable to assume total reflection from the vascular walls. Either this or complete absorption appear the only way to explain the absence of radiation from the sides of the sunflower cotyledons.

Very little experimentation has been done with other plant tissues, and though mitogenetic radiation started with plant tissues, we know much more about radiations from animals than from plants.

The pulp of turnips radiates when 24 hours old (ANNA GURWITSCH); the pulp from *Sedum* leaves does not radiate when fresh, but after 18 hours, it begins and continues until 24 hours old. After 48 hours, radiation has disappeared (GURWITSCH, 1929). Since neither of these two experiments were carried out aseptically, and no accurate record was made of the number of bacteria and yeasts growing in the pulp, they cannot be considered as exact proofs of radiation. *A priori*, we should expect these crushed tissues to radiate because they must contain oxidase.

The radiation of wounds in plant tissues will be discussed together with the wounds of animals on p. **173**. The plant tumors will be discussed on p. **178**.

So far, the discussion on higher plants has been limited to the *emission* of rays. Just as important is the reaction of plants to mitogenetic rays. There are innumerable data on experiments with onion roots. The facts as well as the interpretations have been discussed in great detail in preceding chapters.

No other part of grown plants or seedlings has ever been tried as detector. We cannot as yet form any opinion about the bearing of mitogenetic radiation to total growth, to the form-controlling factors and to the reproductive mechanism of higher plants.

In the case of mold spores (see p. 80), while they have been shown to react upon mitogenetic rays, they are also senders, and muto-induction effects have been obtained.

C. EGGS AND EMBRYONIC STAGES OF HIGHER ANIMALS

(a) *Eggs as Senders.* The eggs of animals are strong senders as well as good detectors, as far as they have been investigated. **Frank** and **Salkind** (1927) observed that with eggs of the arctic sea urchin *Strongylocentrotus Dröbachensis,* radiation does not begin immediately after fertilization; it occurs approximately 1 hour later, and continues for about 1 hour (Table 45). At this time, the amphiaster stage is reached. The first cleavage furrow appears after 2 hours and 45 minutes. For 1 hour before and for 30 minutes after the first cell division, there is no noticeable emission of rays. Half an hour after the first division, radiation begins again.

Warburg (1909), experimenting with the Mediterranean species *Strongylocentrotus lividus* which produces the first cleavage furrow in 40 minutes, had observed a large increase in the rate of respiration of the egg 10 minutes after fertilization. These 10 minutes would correspond to about 40 minutes in the arctic species. The increase in oxygen consumption begins about 20—30 minutes earlier than radiation. **Herlant** (1918) observed a great increase in permeability 2 minutes after fertilization. **Gurwitsch** (1932, p. 99) assumes that a prophase of the "mitotase" diffuses from the egg plasma, becomes activated on the egg surface, and acts upon the "mitotin" which also diffuses out. The chemical reaction furnishing the radiant energy would thus take place on the egg surface, and not within the egg.

(b) *Eggs as Detectors*: Fertilized eggs not only send out rays, but also respond to radiation. **Maxia** showed in 1929 that sea urchin eggs can be stimulated in their rate of development by mitogenetic rays. **Zirpolo**'s data (1930) on the same subject

Table 45. Effect of Sea Urchin Eggs at Different Stages after Fertilization upon Onion Roots

Time after Fertilization	Number of Mitoses Control	Number of Mitoses Exposed	Induction Effect
0 to 1 hr + 10 min . .	167	173	+ 0.3
	299	299	0
0 to 1 hr + 45 min . .	139	203	+46.0
33 min to 1 hr + 50 min . .	215	301	+40.0
1 hr + 10 min to 2 hrs + min 15 . .	67	109	+62.0
1 hr + 45 min to 3 hrs	214	226	+ 0.5
	182	186	+ 0.2
1 hr + 50 min to 3 hrs + 15 min . .	92	86	− 0.6
2 hrs + 15 min to 3 hrs + 15 min . .	268	268	0
	144	157	+ 0.9
0 to 2 hrs + 45 min . .	423	568	+34.0
	252	340	+35.0

are given in Table 27 p. 82. Salkind, Potozky and Zoglina (1930) proved the same for the eggs of the protoannelids *Saccocirrus papillocercus* and *Protodrilus bobrezkii*. The eggs emitted mitogenetic rays during their development, and *vice versa*, their development was accelerated by the rays from isolated frog hearts, crab hearts or the hemolymph of crabs. After an exposure to these radiations for 5 to 10 minutes, a greater percentage of furrowed eggs was observed in 23 out of 26 experiments. The mutual stimulation of sea urchin eggs has already been mentioned on p. 137, as example of allelocatalysis.

According to Wolff and Ras (1934 b), the eggs of *Drosophila melanogaster* are good detectors. They were exposed for 15 to 30 minutes to a broth culture of *Staphylococcus aureus*, 3 hours old, in quartz tubes, and the percentage of hatching eggs was ascertained in equal time intervals. Table 45a shows that at the same moment, always more irradiated eggs had hatched than unirradiated ones.

(c) Embryonic and Larval Stages as Senders: The most detailed earlier investigation of this kind is that by Anikin (1926) with the embryos of the axolotl (*Ambystoma tigrinum*). From very young embryos, with open medullar furrows, the medullar plates were dissected, ground to pulp and used as sender.

Table 45 a. Effect of a Culture of *Staphylococcus aureus* upon the Rate of Hatching of the Eggs of Drosophila

Controls			Irradiated Eggs				Percentage Increase over Control
No. of eggs	No. hatched	Per cent hatched	No. of eggs	No. hatched	Per cent hatched	Time of irradiation	
39	25	64	51	45	88	15—20 min.	24 ± 10.6
81	15	18.6	72	30	41.7	15—20	23.1 ± 7.45
52	18	34.6	60	49	81.7	15—30	47.1 ± 8.1
304	210	69	312	300	96.1	20	27.1 ± 8.5
324	147	45.4	304	228	75	20	29.6 ± 3.7
344	255	74	327	323	98.4	20	24.4 ± 2,6
366	136	37	357	244	68	20	31 ± 3.5
118	79	67	117	98	83.5	20	16.5 ± 5.5
74	38	51.3	85	60	70.6	20	19.3 ± 7.7
1702	923	54.2 ±1.2	1685	1377	81.7 ±0.95		27.5 ± 1.54

The same was done with the rest of the embryo. As detector, onion roots were used. The results are shown in Table 46. Only the medullar plate radiated. Then, living embryos, after the removal of the surrounding mucus, were placed in a glass tube so that either only the ventral or only the dorsal side faced the detector root. Even at the morula stage, the vegetative hemisphere did not radiate. During gastrulation, the entire blastopore seems to radiate.

With slightly larger embryos, just previous to hatching, the brain could be dissected out, and it was found that the brain, but none of the other tissues, radiated. The brain pulp of the fullgrown animal did not (see however p. 158).

This agrees with the earlier statements of GURWITSCH, and of REITER and GABOR, that frog tadpoles radiate only until about 1 cm. long, and that the center of radiation appears to be in the head. Concerning the radiation of certain organs during metamorphosis, see p. 167.

Among the invertebrates, the embryos of *Saccocirrus* radiate during the entire stage of their development. SALKIND (1931) found the following induction effects:

Blastula Stage	30%	50%	48%	34% induction
Gastrula Stage	63%	30%	30%	
Swimming Larvae . .	54%	37%	49%	32%
Trochophore Stage . .	37%	28%		

Table 46. Effect of various parts of the embryos of the axolotl upon onion roots

Embryonic Stage	Number of Mitoses Control	Number of Mitoses Exposed	Induction Effect %
living morula stage, animal hemisphere	750	931	+24
living morula stage, vegetative hemisphere	428	410	— 4
living embryo, before hatching; dorsal side	230	268	+16
living embryo, before hatching; ventral side	492	471	— 4
pulp of medullar plate	324	409	+26
pulp of embryo without medullar plate	482	505	+ 5
pulp of embryos' brains I	627	832	+33
pulp of embryos' brains II	865	1042	+20
pulp of embryos' brains III	920	1076	+17
pulp of entire embryo without brain I	724	728	0
pulp of entire embryo without brain II	689	696	+ 1
pulp of embryo's liver	631	635	+ 1
pulp of brain of grown animal I	617	632	+ 2
pulp of brain of grown animal II	848	847	0

Of the insects, only the larvae of *Drosophila* have been analyzed. They do not begin to radiate until shortly before pupation, and cease to radiate 90 hours after this (see also p. 169).

Quite different is the behavior of the chicken embryo. Sorin and Kisliak-Statkewitsch (1928) working with entire embryos two days old and also testing the brains of older embryos could find no evidence of radiation. However, during the second and third day of incubation, positive results were obtained with the liquefied zone around the embryo.

(d) Embryos as Detectors. The only example of embryos or larvae as detectors of mitogenetic radiation is that of Blacher and associates who could make the fore leg of a tadpole grow more rapidly by exposure to radiation (see p. 168). The morphological changes in sea urchin larvae observed by Magrou may also be mentioned here (see p. 165).

D. TISSUES OF ADULT ANIMALS

Tissues as Senders: Until recently, most of the tissues of adult animals had been thought to be non-radiating. If we consider radiation to be produced largely by the chemical reactions in the cells, it would seem that all tissues should radiate quite strongly. **Gurwitsch** (1934) has given some convincing explanations for the differences between radiating and non-radiating tissues.

The very strong absorption of these short-waved rays has already been emphasized repeatedly. **Gurwitsch** found that extremely thin films of oil or related substances, films of practically only one molecule thickness, suffice to absorb completely a mitogenetic radiation of normal intensity. On the other hand, equally thin films of substances capable of enzymatic cleavage, such as lecithin, are also capable of secondary radiation (see p. 45), and they will pass on radiation not as a beam, but in all dimensions of the film. Thus we observe strong radiation in blood which contains no membrane, and in nerves which contain plenty of lecithin. These two will be treated separately in the next two chapters, on account of their importance.

The cornea of the eye is also a good sender, being a continually renewed tissue. **Wolff** and **Ras** (1933*c*) consider it to be one of the strongest sources, about 10 times as strong as blood. **Gurwitsch** has recently favored the peptic digestion as a strong and fairly constant source. Other actively reacting digestive enzymes are likely to be good senders, but would hardly be considered as tissues. **Potozky**, **Salkind** and **Zoglina** (1930) studied the tissues of two crabs, *Carcinus maenas* and a species of *Pachygrapsus*; they found the pulp from gills and from testicles inactive, while the hepatopancreas radiated distinctly. This organ is the seat of active proteolysis. Probably, autolyzing muscle would radiate.

With vertebrates, negative results had been obtained by all earlier workers with lymph glands, testicles, ovaries, skin, liver, and with the resting muscle. The working muscle, however, proved to be very active. Negative results, particularly when obtained with tissue pulp, are now considered of little significance since better methods, especially with organs *in situ*, showed distinct radiation.

On p. 65, SIEBERT's results have been given, showing that the muscle radiates only during work, and that pulp from resting muscles does not radiate, while that from active, tired muscle does. According to recent experiments by FRANK and KREPS (quoted from GURWITSCH, 1932, p. 149), this result was caused by an irritation of the muscle during grinding. By dropping the organ into liquid air and grinding it while frozen, the results were the opposite. In pulp from working muscle, the pH changed from 5.96 to 5.82 during the experiment, and radiation was absent; while with rested muscle, the pH changed from 6.33 to 5.85, and radiation was present. This refers only to the pulp, however; the muscle *in situ* radiates strongly while working, but weakly also when resting.

The spectral analysis indicates that the main source of muscle radiation is not glycolysis; partly it is an oxidation process, partly of unknown origin. It must be remembered that there is no quantitative relation between the total energy liberated by a chemical reaction, and the ultraviolet radiation emitted (see p. 41). It may well be that some proteolytic process whose energy output is negligible for the muscle work emits most of its energy in radiant form, while glycolysis, with a much larger total energy output, emits only a very small fraction of it as mitogenetic rays.

A good example of applied spectral analysis should be mentioned in this connection. GURWITSCH had been greatly puzzled in his earlier work by the observation that the very distinct radiation from the rabbit's eye disappeared during starvation, but reappeared after 8 days of continuous starving. Several years later, spectral analysis brought a simple explanation (GURWITSCH 1932, p. 67). The normal radiation is glycolytic, and ceases during starvation because of lack of sugar. After prolonged starvation, proteolysis of the tissues becomes necessary for the continuation of life processes, and this produces a proteolytic spectrum.

Tissues as Detectors: Only a few instances are known where fullgrown animals or their tissues reacted upon mitogenetic rays. The role of radiation in wounds will be treated in one of the following chapters.

With multicellular organisms, the stimulation of the growth-rate is not so readily proved. In growing organs, the cells lie so closely together that the optimal intensity of radiation may already be furnished by the organ itself. Emanations from outside, if they are not absorbed completely before they reach the region of growth, very likely can only be harmful.

A few instances are known, however, where the rate of cell division is accelerated. The onion root is usually considered the classical example though the data have been interpreted somewhat differently by **Reiter** and **Gabor** (see p. 129). Another example is the cornea (see p. 84). Further illustrations are the effects of resorbed tissue in different stages of amphibian metamorphosis (see p. 167).

These latter results together with those obtained with embryos (p. 144) suggest very strongly that the developmental mechanism controlling size and form makes use of ultraviolet radiations as well as of purely chemical means to achieve its purpose. This and the possibility that neoplasms, especially cancers arise through mitogenetic radiation will be discussed in later chapters.

The only animal tissue that has been actually used as detector is the cornea. According to **Gurwitsch**, it yields very good results (see p. 84).

E. BLOOD RADIATION

A comprehensive review of all work on blood radiation has been given by W. **Siebert** (1934) in the second volume of „Handbuch der allgemeinen Hämatologie".

Blood radiates quite strongly, even in adult animals and men, the only exception being extreme old age and a very few diseases which will be discussed later. The blood of various mammals, birds and amphibia, and also the hemolymph of the crabs *Carcinus* and *Pachygrabsus* and of the clam *Mytilus edulis* has been tested, and strong mitogenetic effects have always been observed. According to **Kannegiesser** and **Kazwa** (quoted from **Gurwitsch**, 1932, p. 124), dog's blood which possesses only very weak glycolysis gives very fluctuating results.

Blood radiates within the veins or arteries as could be shown by removing the tissues around the veins or arteries, and exposing a detector to the blood radiation through the inner wall of the blood vessels which is transparent to these rays (spectrum see fig. 44).

Outside the blood vessel, blood loses its radiating power within 10 to 15 minutes; this holds also for the hemolymph of crabs. In hemolyzed blood, radiation can be restored by adding glucose.

For radiation experiments, blood is mixed with an equal amount of a 4% $MgSO_4$ solution to prevent clotting, or it is hemolyzed with distilled water (POTOZKY and ZOGLINA, 1929). It is possible to dry blood on filter paper and thus preserve its radiating power for 2 to 3 days. Since this might be of practical importance, especially in diagnostic medicine, GURWITSCH's method (1932, p. 121) shall be mentioned here:

> The blood should be drawn without previous disinfection of the skin (iodine, alcohol) because traces of disinfectants appear to inhibit glycolysis. The drops are spread widely on the filter paper to prevent coagulation and thick layers. Drying must be accomplished very rapidly to preserve full radiating power; the dried sample should be kept dark. In making the test, detector and everything else must be prepared before water is put upon the dry blood. The spot is cut into very small pieces, and softened in a tiny shallow dish with 5 to 6 drops of distilled water, under continuous stirring. As soon as the water becomes dark red, it is drawn off with a pipette and used at once for irradiation. From the adding of the water to the beginning of exposure, not more than 1 to 1.5 minutes should elapse.

HEINEMANN's method for detecting blood radiation has been mentioned on p. 72.

With starving mammals, the blood does not radiate, but can be brought back by adding glucose. POTOZKY and ZOGLINA starved rats until they had lost about 30% of their body weight. The mitogenetic effects obtained with the blood of these animals were

without addition	—2.5	—5	—2	0
with 2% glucose	21	44	36	39

These strong effects could be obtained only by adding much more sugar than is normally contained in the blood.

A very important observation is the disappearance of blood radiation after continued work. BRAINESS (quoted from GURWITSCH, 1932, p. 132) investigated a number of laborers before and after the day's work. Table 47 gives a few of his data which show consistently no radiation immediately after long-continued work, but normal radiation after 2 hours of rest. This must be considered when making blood tests for diagnostic purposes.

Table 47. Mitogenetic Effects from the Blood of Factory Workers before and after Work

Person	Month	before work	at the end of the day's work	2 hours later
K	October	25	6	—
K	January	24	— 1	36
K	December	45	1	26
B	November	38	— 3	31
B	January	28	6	22
M	January	32	— 5	11
E	January	33	2	27
S	January	35	—13	10
S	January	21	12	27
D	January	26	1	30

Wassilieff (1934) repeated the investigation with mental work (calculation). The first results showed a decrease in radiation, but this happened before psychic analysis showed mental fatigue, and it could be demonstrated that muscular work connected with calculation caused the decrease in radiation while the mental work as such does not affect it.

The spectrum of the radiation of rabbit blood from the streaming blood in the *vena saphena* is shown in fig. 44 as measured by Golischewa (1933). It has practically all the lines characteristic for glycolysis, proteolysis, creatin hydrolysis, phosphatid hydrolysis, and oxidation, and, in addition, some new lines. Of these latter, the lines 1920—30, 1940—50, 1960—70 and 2000—2010 have also been found in nerve spectra (fig. 47).

The main source of blood radiation with mammals appears to be glycolysis. In addition to the starvation experiments just mentioned, Kannegiesser and Kawza made extensive experiments with dog's blood, measuring the glycolytic power and the radiation of each sample. There was a good, though not quantitative, relation between the two. Intravenous insulin injection reduced glycolysis and radiation to zero. In some cases, addition of glucose (probably in overdose) interfered with radiation.

Rat blood treated with heparin loses its radiation by NaF, but not by KCN. This suggests glycolysis, which is affected by

NaF, but not by KCN. Hemolymph of crabs, however, loses its power promptly in the presence of 0.0001 molar KCN, and so does hemolyzed rat blood. This suggests that in normal rat blood, glycolysis is the dominant source, but in hemolyzed blood, and in the blood of crabs (which contains no hemoglobin) oxidation is the deciding reaction.

Of the various fractions of the blood, the serum radiates immediately after the blood is taken from the animal, but loses this power rapidly. It continues, however, to produce secondary radiation, even after several months (p. **43**). The leucocytes, polynuclear as well as lymphocytes, radiate distinctly but not very strongly, according to Klenitzky (1932). The spectrum indicates glycolysis, oxidation and phosphatase action, and probably proteolysis.

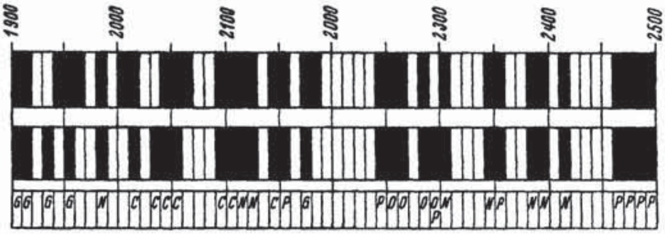
1900
2000
2100
2000
2300
2400
2500
G G G G N C C C C C C N N C P G P O O O O N P N P N N N P P P P

Figure 44. Above: spectrum of streaming blood in the artery of a rabbit. Below: the combined spectrum of 5 common biological processes (oxidation = *O*, glycolysis = *G*, creatinin hydrolysis = *C*, proteolysis = *N*, phosphatid hydrolysis = *P*).

The intensity of blood radiation is greatly changed after wounding (see p. **174**). During the metamorphosis of amphibia, it varies enormously in the different stages of development.

Protti (1931) observed a great decline or complete absence of radiation in blood during senility, and this result was confirmed by Heinemann (1932). In consequence Protti injected blood from young persons into old ones. Distinct clinical evidence of "rejuvenation" by this treatment has been claimed, and the blood of old people thus treated radiated again. Table 48 gives some of Protti's results.

A somewhat different way was used by Heinemann and Seyderhelm to renew blood radiation in old persons. In his effort to find the reason why ultraviolet light was beneficial to animals, Seyderhelm (1932) succeeded in isolating a compound

Table 48. Mitogenetic Effects Produced by the Blood of Old Persons After Injection of Blood from Young Persons

Blood Group	Mitogenetic Effects			Clinical Results
	young blood	old blood	old blood after injection	
A	70	5	30	very good
A	20	3	4	none
A	60	29	38	good
A	70	15	38	very good
A	20	10	10	little
A	80	30	53	very good
B	72	16	32	good
O	81	24	40	very good
O	62	30	37	good
O	22	17	20	little

from the blood corpuscles which he called *cytagenin*. When this substance was injected into healthy persons, it increased blood radiation (fig. 45). With patients suffering from anemia, it washed the newly-formed blood corpuscles out of the bone marrow and produced a normal blood picture (HEINEMANN, 1932). This was possible in all cases of secondary anemia, but not in pernicious anemia in which the bone marrow no longer produces blood cells.

When injected into patients 70 to 80 years old, without blood radiation, cytagenin produced either no effect or a slight depression effect during the first days, but after 1 to 2 weeks, the blood began to radiate, and repeated tests showed no decrease. How long these experiments were continued, HEINEMANN did not state.

The blood of asphyxiated animals does not radiate. It loses this power before the animal is dead, even when the process is still reversible.

Quite remarkable is the persistence of blood radiation during illness. L. GURWITSCH and SALKIND (1929) obtained radiation from the blood of tuberculous guinea pigs until almost immediately before death. Diabetes, lues, osteomyelitis and ulcer of the stomach did not decrease blood radiation. The only diseases which showed this conspicuous absence were leucemia, and severe septicemia with high fever (also poisoning with nitro-benzene),

and cancer. This was verified by SIEBERT who observed absence of radiation in severe cases of sepsis, pneumonia, and scarlatina, and by GESENIUS (1930). HEINEMANN (1932) added chronic tonsilitis to this non-radiating group; in some cases, radiation appeared again soon after the removal of the tonsils.

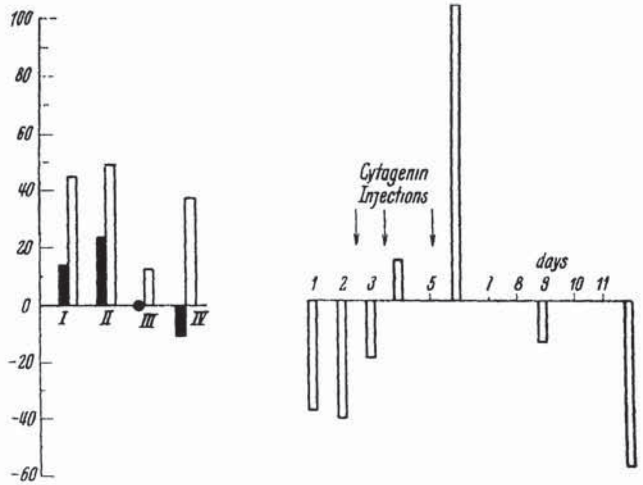

Figure 45. Increase of blood radiation by injection of cytagenin. At left: black indicates radiation before injection, white after injection. *I* and *II* are normal healthy persons, *III* is a very old person, *IV* is a carcinoma patient. — At right: the effect of repeated injections upon a carcinoma patient.

Fig. 46 shows the results obtained by GESENIUS (1930) who measured blood radiation by the decrease of yeast respiration (see p. 84). Since only very few easily recognized diseases prevent blood radiation, it can be used for the diagnosis of cancer (see p. 180).

A very comprehensive review of his extended research on blood radiation has been given by PROTTI (1934a). All measurements refer to apparently healthy persons, and are

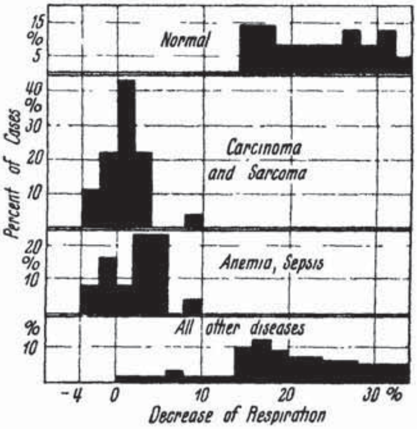

Figure 46. Decrease of respiration of yeast irradiated with the blood of healthy and sick persons.

made by the yeast bud method by means of the hemoradiometer (p. 66). In a large number of tables and graphs, he demonstrates the decrease of radiation with age, the stronger radiation of tall, slender people as compared with short individuals with a tendency for stoutness, and the slight increase of male over female blood. In the same individual, radiation increases 1 to 2 hours after each meal, and decreases with physical fatigue. During the winter months, it is slightly lower. It is increased by high altitudes, also by a trip to the sea shore, further by inhalation of oxygen, though in this latter case, the effect does not last more than one hour. Menstruation and pregnancy have characteristic curves.

Protti states that even among healthy individuals, "normoradiant, hypo-radiant and hyper-radiant" types can be distinguished, all reacting in a parallel manner upon the same changes.

F. NERVE RADIATION AND THE CONDUCTION OF STIMULI IN ORGANISMS

The results obtained with secondary radiation led Gurwitsch to the idea that possibly secondary radiation might be a controlling factor in the conduction of stimuli in plants and animals. Let us recapitulate briefly the facts obtained with onion roots (see p. 140).

Irradiation of the older, upper part of the severed root produced radiation from the tip. Irradiation of the tip produced radiation from the older tissue. The impulse was conducted longitudinally at the rate of approximately 30 meters per second, but there was no conduction to the diametrically opposite side of the root.

Later, Frank observed that the same held true for muscles. The resting sartorius muscle of a frog when irradiated biologically at a definite place for 10 seconds emitted a strong mitogenetic radiation from a place 20 mm. away. In fact, at a distance of 20 mm., the effect was much stronger than at 10 mm.; the intensity had increased by conduction.

These results, together with the discovery of secondary radiation in nerve tissue, inspired in Gurwitsch the bold thought that conduction of stimuli in nerves and muscles may be accomplished or aided by secondary mitogenetic radiation. This statement is possibly too blunt, but it expresses in a few words the ultimate aim. GURWITSCH himself (1932a) says that this theory may appear bold, but it seems justified to approach it experimentally, on account of several suggestive facts.

One of them is the possibility of studying, by means of the mitogenetic spectrum, the metabolism of the nerve in the resting and in the excited stage. It is evident that the chemical analysis can give only a rather incomplete picture of the chemistry of nerve stimulation, because it involves destruction of the nerve, while radiation can be observed without injury. The result of the spectral analysis of nerve radiation has, as a matter of fact, revealed some chemical processes which had not as yet been discovered by chemical analysis.

The views concerning nerve radiation have undergone considerable change with the improvement of the methods. The older experiments by ANIKIN with the brain of adult salamanders (see p. **145**) and those by REITER and GABOR (1928) with the sciatic nerve of the frog gave negative results. They were confirmed by WASSILIEW, FRANK and GOLDENBERG (1931) who obtained positive results only with the olfactory nerve of the pickerel. By using the yeast volume (p. **73**) as detector, KALENDAROFF (1932) not only proved that the sciatic nerve of the frog radiated distinctly, but he could also study its spectrum by using intermittent exposure.

The spectrum was determined with resting as well as with irritated nerves. Irritation of the nerve was accomplished either by cutting into it (traumatisation), or by electrical tetanization with platinum electrodes 3—4 mm. apart, 8—12 shocks per minute (faradisation), or mechanically, by hitting with a light hammer, intermittently. All experiments were conducted with intermittent exposure by the rotating disk (see p. **105**). This not only increased the intensity of the effect, but also eliminated the possibility of secondary radiation from the nerve induced by the yeast detector.

The minimal total exposure requred to bring about mitogenetic effects was approximately 6 minutes with the resting nerve and 2—3 minutes with the excited one. The detailed spectra are shown in fig. 47. The rather surprising fact was revealed that different kinds of irritation produce slightly different spectra.

Further, it was found that radiation at the point of irritation differs from that of the same nerve a short distance away.

The spectra (of which each 10 Å strip is the result of at least three determinations) show five well-known chemical processes: oxidation, glycolysis, phosphatase action, cleavage of creatinin phosphate, and de-aminisation of amino acid A few lines were observed also which cannot at present be accounted for.1)

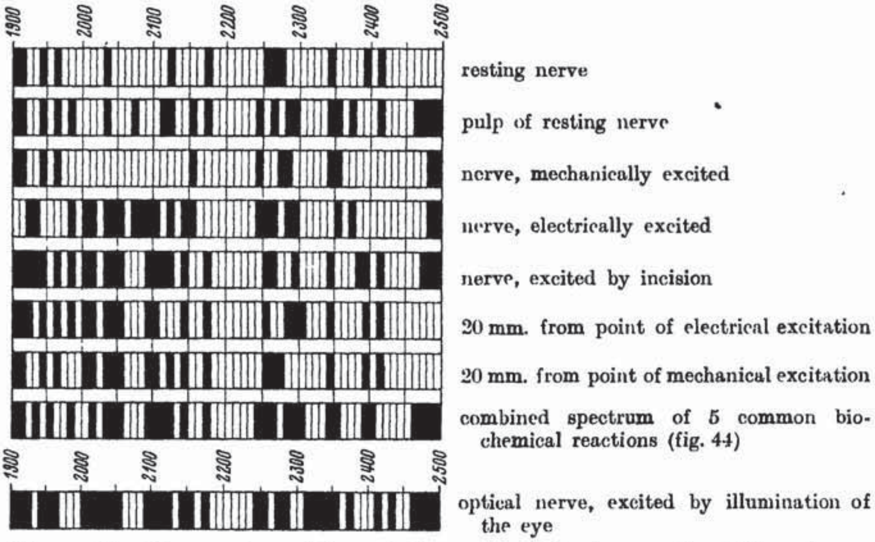

Figure 47. The spectra of the sciatic nerve of the frog under different conditions. The lowest line represents radiation from the optical nerve *in situ.*

These five chemical processes agree with chemical investigations on nerve metabolism. Strange is the absence of certain lines. In mechanical and electric irritation, most of those concerned with de-aminisation are missing, and in the electrically-stimulated nerve, the glycolytic regions are absent while in the spectrum of the same nerve, 20 mm. removed from the zone of

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1) Two lines of the glycolytic spectrum, 1930—40 and 1950—60, are missing in most of the nerve spectra while they all have the neighboring lines 1940—50 and 1960—70. This may be due to a slight shift in the calibration of the instrument. Line 2410—20 might also be accounted for in this way, as a shift of the preceding line to the right, or rather an error in the calibration to the left.

However, the line 2000—10 in more than half of the spectra and 2370—80 in 3 of the spectra cannot be explained by the same error of calibration.

irritation, they are present. The creatinin lines are absent in mechanical stimulation. The spectra of nerve conduction lack entirely the phosphatase lines.

This difference in the spectra raised the question of "adequate" nerve stimulation. A good example of this has been furnished by ANNA GURWITSCH (1932) with the optic nerve of the frog. After decapitation and removal of the lower jaw, the optic nerve was laid bare by dissection from the roof of the mouth. A small window, 1.5×2 mm., is sufficient to permit access to the chiasma and tracti optici with part of the optic nerve. The radiation from this was tested in the darkroom while directing a beam of light on one of the eyes.

Increase in number of yeast cells by direct microscopic count was the method used to prove the radiation. The results were always positive while controls without illumination of the eye showed consistently no effect.

The spectrum is shown in the lowest line of Figure 47. It agrees essentially with those by KALENDAROFF. It has also some unknown lines agreeing with KALENDAROFF's. Those corresponding to proteolysis (de-aminisation) are present only in part.

These experiments have revealed that there are differences in the metabolism of the resting and the excited nerve, and that even the type of excitation may change the spectrum. It might be well to remember here GURWITSCH's statement that these spectra represent "minimal spectra". The established lines are doubtless correct, but there may be other lines too weak to be recorded by the detectors used.

These fine results induced ANNA GURWITSCH (1934a) to approach the more fundamental question whether this radiation of the optic nerve after illumination of the eye was only a simple secondary radiation, or represented an important functional part of nerve conduction. She laid bare the optic nerve, the optic lobes, the medulla oblongata and the spinal cord of living frogs, and kept these parts covered with the skin. For several hours after the operation, all nerves radiated strongly, but 24 hours later, lobes and hemispheres did not radiate while medulla and spinal cord usually continued to radiate weakly. As soon as light was directed onto the eyes of the frog, the lobes and hemispheres radiated strongly while the medulla and spinal cord did not change.

When the optic lobes were still in the excited stage from the operation, they did not react promptly upon illumination of the eye. This suggested an interference between traumatic radiation and normal reaction upon optic irritation. The explanation could be verified by irritating electrically the sciatic nerve. Before the circuit was closed, the lobes reacted strongly upon illumination of the eye; when the current was applied, the reaction upon light was weak and irregular; after ceasing the irritation, but continuing illumination, radiation ceased completely for a short time, and after that, the normal strong effect appeared.

It had been shown at this time (p. 45) that secondary radiation usually is a resonant radiation responding only to the wave lengths characteristic for it. ANNA GURWITSCH studied the reactions produced by various spectra applied to the chiasma of the optic system. The emission from an electrically irritated nerve made the lobes radiate weakly, while the hemispheres showed no effect. Yeast radiation, however, induced strong radiation in both. Addition of the glycolytic component (1900—1920 Å from a monochromator) to nerve radiation produced also good radiation in lobes and hemispheres.

The spectrum of the radiation of the lobes contains always glycolytic lines even when radiation has been brought about by exposure of the chiasma to rays from the oxidation of pyrogallic acid which are very different from those of glycolysis. It is, therefore, not a mere secondary radiation, but indicates the release of an unknown independant chemical reaction in the optic lobes.

The extent of radiation in a nervous system during illumination permits thus the experimental approach to the problem of localisation of functions in the brain.

In another paper, ANNA GURWITSCH (1934 b) obtained different spectra from the hemispheres of the frog brain when the eyes were illuminated with different colors. Green light produced the lines of glycolysis, proteolysis, oxidation and phosphate cleavage. With red light, the last mentioned part was missing, and with blue light, there were no proteolytic lines. Probably, the metabolism of the nerve was not changed completely, and all lines were present, but their relative intensity was varied.

The intensity of the colored lights used and the wave lengths of the colors are not mentioned.

The effect of the intensity of light was tested by **Anna Gurwitsch** (1932) by varying the distance between light and eye from 6 to 18 cm. (i. e. varying the intensity from 9 to 1) and determining the necessary exposure time. The result was as follows:

Length of Exposure . .	1	3	5	10	15	20	25	30	35 seconds
Induction Effect at 6 cm.	5	19	22.5	4	—	—	—	—	—
Induction Effect at 18 cm.	—	1	4	--	4	4	13	16	6

The weaker light (18 cm. distance) requires a longer time to produce the effect, but there is no appreciable difference in the *intensity* of the effect.

It could further be shown that continued exposure to light produces a long after-effect if the head is cut from the animal; in the living animal, however, radiation ceases when the eye is darkened (see Table 49).

Table 49. Mitogenetic Effect of the Optic Nerve after illumination and darkening of the eye

	Seconds of Exposure	Mitogenetic Effect		
		dark	light	second darkening
Isolated Head	20	2	22.0	21.8
	20	2	14.5	12.0
	20	—6	20.1	14.6
Living Animal	15	—4.5	22.5	—2.0
	10	0	15.5	—2.5

Another important advancement in the effort to correlate nerve conduction with mitogenetic radiation is the accurate measurement of the velocity of progress of secondary radiation in the sciatic nerve. **Latmanisowa** (1932) found this to be 30±3 meters per second (see p. **112**) and this agrees, within the limits of error, with the rate of conduction of nerve stimuli.

Very suggestive, though not positive proof, are also the experiments by **Latmanisowa** on the "fatigue" of the nerve by continued irradiation with strong light (see p. **111**).

Later publications by **Latmanisowa** (1933, 1934) have added another impressive fact to prove the role of mitogenetic rays in nerve conduction. By exposing the sciatic nerve, with the attached muscle, in a moist chamber for about 2 hours to the radiation from protein digestion, the nerve showed all symptoms of parabiosis.

Two electrodes touched the nerve, and the place between them was irradiated. For about 2 hours, the nerve reacted normally upon electric impulses, causing a stronger muscle contraction with a stronger impulse. After this time, the nerve became at first more excitable, but soon the muscle reactions became weaker and weaker, and then, the typical parabiotic stage set in, the contraction being stronger when the impulse was weaker. Finally, the nerve ceased to react altogether, and it took two hours after the removal of the mitogenetic source before the nerve had recovered, and its reactions became normal again.

This experiment has been repeated more than 40 times with the same success. Controls with gastric juice without protein were not affected. The nerve outside of the irradiated zone reacted normally at the same time when the exposed part of the same nerve showed parabiosis. The author quotes a paper by LAPITZKI who obtained the same effect by rays from a mercury vapor lamp in a few minutes.

The nerve at the parabiotic stage has not ceased to radiate; on the contrary, the emission seems to be much stronger than that of the normal nerve.

It has not been possible as yet to produce muscle contraction, or a corresponding effect, by irradiating a nerve. This may be due to the absence of an "adequate" stimulation. Perhaps, a combination of definite wave lengths is necessary to produce such effects. The one example of true adequate stimulation are the above-mentioned experiments by ANNA GURWITSCH with the optic nerve.

It seems too early to speculate on the relation between the mitogenetic radiation of nerves and the action current. Though a number of physiologists oppose this idea, it does not seem impossible that the two observed facts may be some time be combined to produce a more comprehensive explanation of the nerve mechanism.

G. MORPHOLOGICAL EFFECTS

While the classical example of morphological effects produced by biological radiation is that of sea urchin larvae, they shall be preceded, for the sake of logical arrangement, by a short note on unicellular organisms.

1. Yeasts and Bacteria: CHRISTIANSEN (1928) observed striking morphological changes in yeast cells as well as bacteria under the influence of radiations emanating from menstrual blood. It seems that most of these experiments were carried out without exclusion of chemical effects from volatile substances of the blood, but the same effects could be obtained when a quartz coverglass protected the test organisms perfectly against vapors from the blood or from outside.

At times, the blood was strong enough to kill microorganisms; more commonly, it affected their cell forms. With *Bacterium coli,* the non-motile cells immediately above the drop of blood were 3 to 5 times as long as the farther removed cells; *Bacterium vulgare* lost its pellicle formation; *Lactobacillus bulgaricus* grew to long threads without cell division; *Oidium* formed no oidia. *Streptococcus lactis* and *cremoris* did not appear to be changed morphologically.

Just as striking were the changes in yeasts. Frequently, retardation of growth was accompanied by an enormous expansion of vacuoles, leaving the protoplasm only as a very thin layer around the cell membrane. In other cases, there was a decided tendency to grow into hyphae. Still other cultures showed formation of giant cells.

Similar morphological changes could be produced by exposure of yeast to saliva of apparently normal persons (FERGUSON, 1932). Particularly the large spherical cells with tremendously extended vacuoles and with complete loss of granulation are considered typical "saliva cells". It has not been possible, however, to produce these same forms by interposing a quartz coverglass between saliva and yeast. Thus, the physical nature of this phenomenon is not proved. On the other hand, it is evidently not a purely chemical effect of some saliva constituent because the yeasts grew quite normally in mixtures of equal volumes of saliva and raisin extract; these cultures must have obtained more saliva constituents than could possibly distil over from the

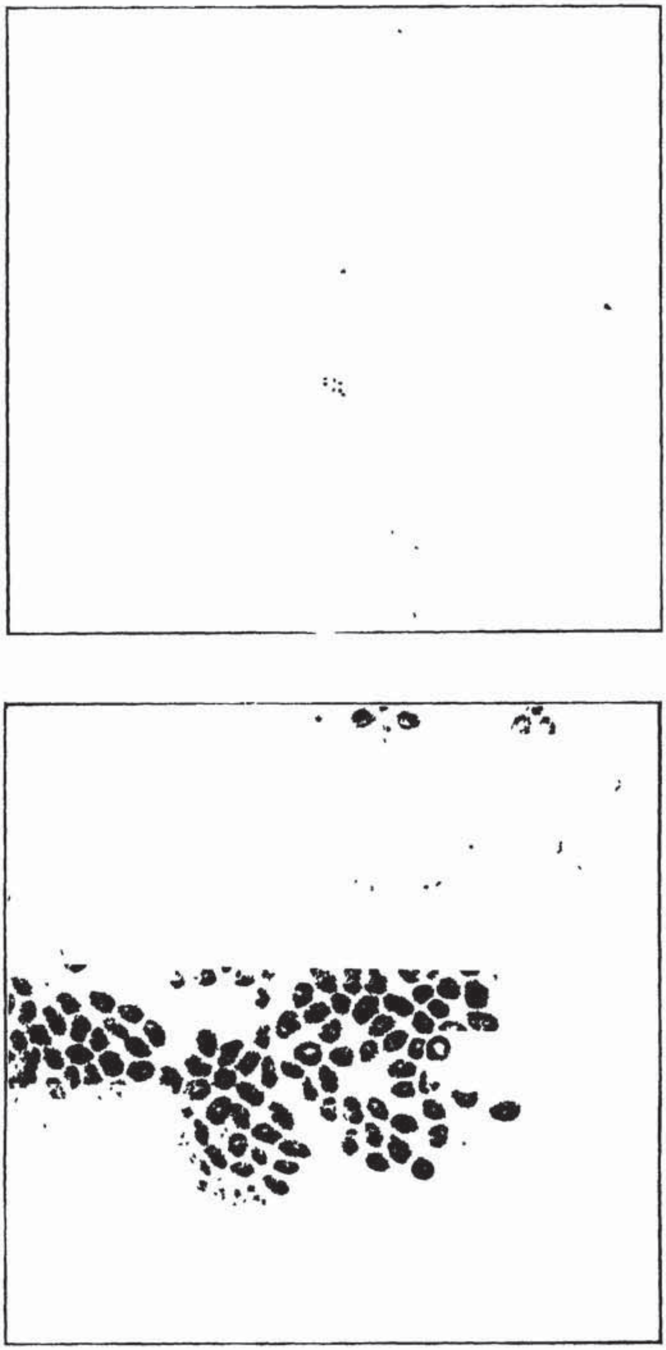

Figure 48 a. Normal growth of *Saccharomyces Mycoderma punctisporu*
Above: 100 times; below: 500 times.

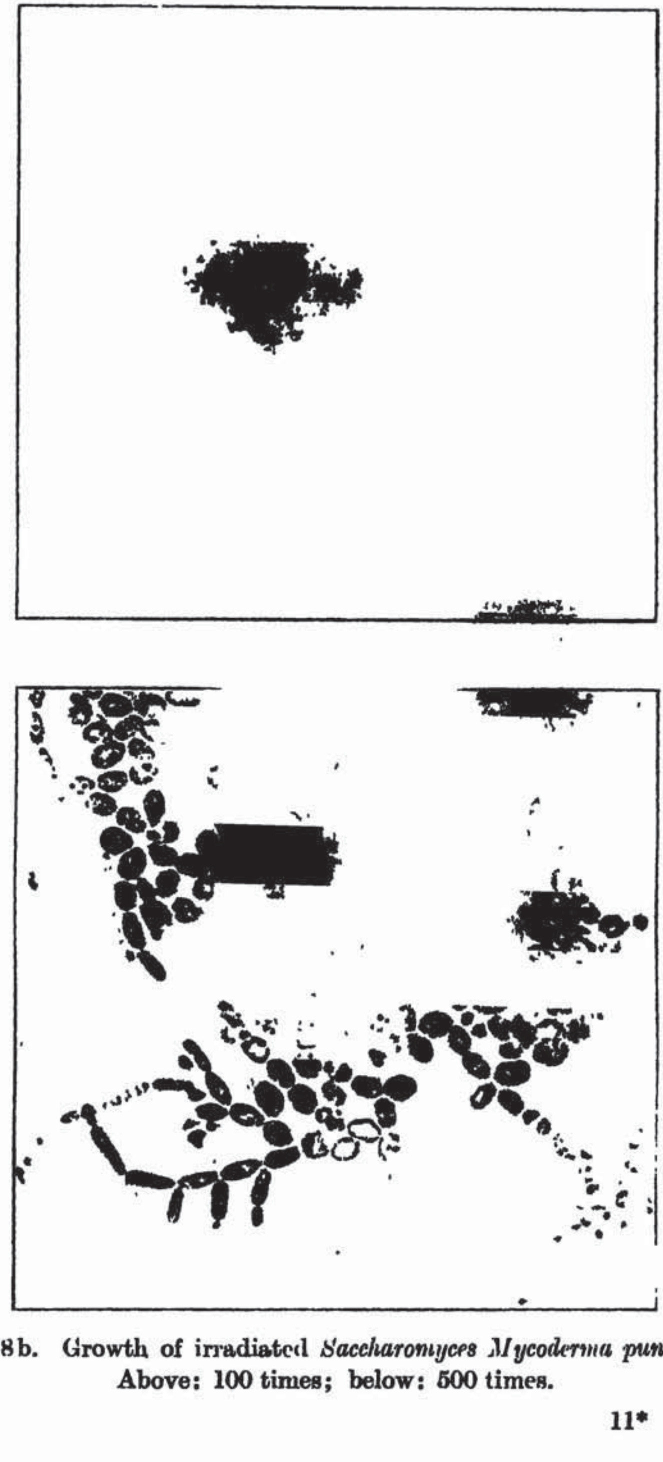

Figure 48b. Growth of irradiated *Saccharomyces Mycoderma punctisporus*. Above: 100 times; below: 500 times.

saliva drop to the yeast culture. These experiments, as well as those by CHRISTIANSEN, were made in hanging droplets on a coverglass which was sealed to a moist chamber slide with vaseline or lanolin; the "sender" was at the bottom of the cavity.

By the same method, the effect of plants upon the morphology of yeasts was studied. It was found that most parts of the various plants stimulated yeast growth considerably, and also produced great morphological changes. The most sensitive yeast was *Saccharomyces Mycoderma punctisporus* GUILLIERMOND which was frequently greatly elongated so that it appeared under the low power like a mold colony. However, true branching was never observed, and the growth resembled more that of a *Monilia* Fig. 48 shows the change produced by exposure to a young agar culture of the same species. Some of the wine yeasts also showed changes of cell form and size.

Seed embryos, pollen, and roots produced the strongest effects; leaves, the weakest effects.

These results could not usually be repeated with the interposition of a quartz coverglass between yeast and sender. Only the one or the other symptom appeared occasionally (see fig. 48), but scarcely ever the complete set of morphological changes. However, the steam distillates of carrots or potatoes, when added to the culture medium in large amounts did not affect the morphology; nor did the juice of crushed carrots produce any change. When the sender was poisoned chemically, killing all cell activity, no effects were observed.

Perhaps we are dealing here with a combined chemical and physical effect, such as STEMPELL (1932) assumed to be rather common in nature.

In the author's laboratory, Mrs. BARNES has isolated a bacillus which through a quartz coverglass, will change morphologically the yeast cells, giving them the appearance of "saliva types".

2. Sea Urchin Larvae. It has already been discussed on p. 86 that sea urchin eggs, when exposed continously to mitogenetic rays from various sources, develop into very abnormal larvae. It has also been stated there that recently, it has been suggested that this is not due to a real radiation, but to an electric effect. Microphotographs of these forms are shown in fig. 49.

J. and M. MAGROU (1931) have studied the abnormal larvae histologically, and have come to the conclusion that they are primarily due to overproduction of the mesenchyme, while the ectoderm and endoderm appear normal (fig. 50). This gives a simple explanation of the morphological changes by the mito-

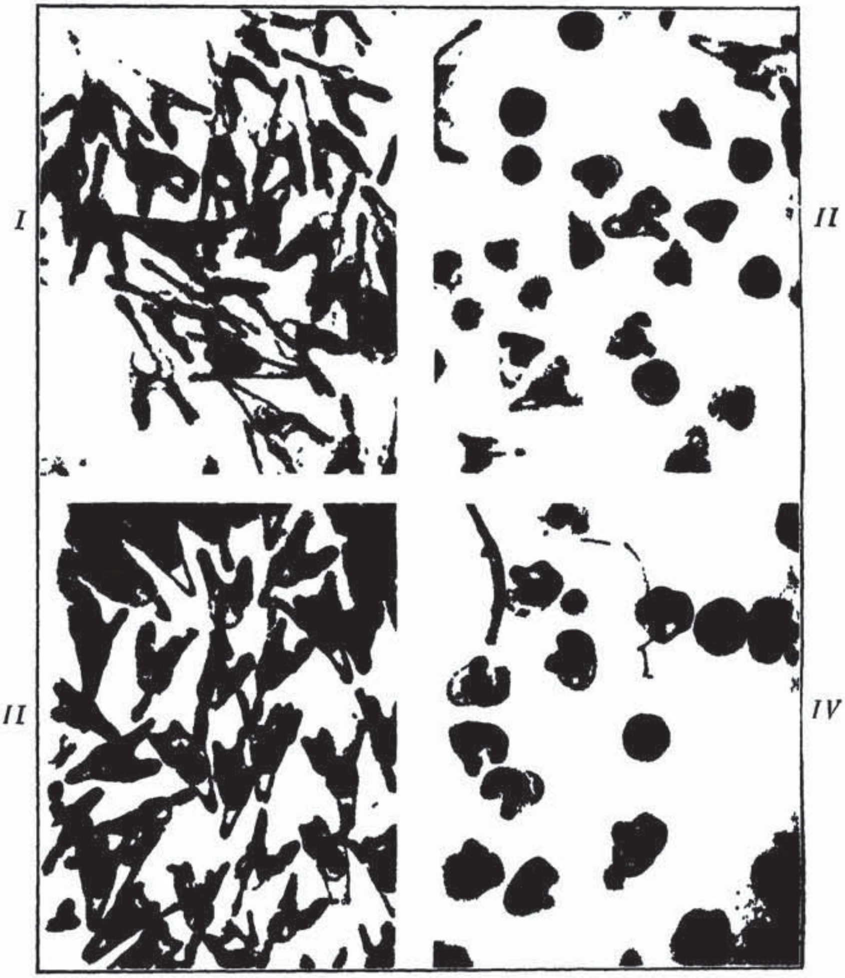

Figure 49. Larvae of the sea urchin. *Paracentrotus lividus.*
I. Control, untreated.
II. Same origin and age as *I*, but exposed continuously through quartz to an acid solution of phenosaffranine reduced by $KHSO_3$.
III. Control, fertilized with normal sperm.
IV. Eggs of the same origin as *III*, but fertilized with sperm exposed for 45 minutes to the same solution as *II*.

genetic effect. However, there is considerable difference in the reaction of ectoderm and endoderm cells on one side, and mesenchymatic cells on the other. The former are not essentially affected (there may be some retardation eventually), while the mesenchyme grows entirely out of bounds though it is really protected against radiation by the outer cell layers.

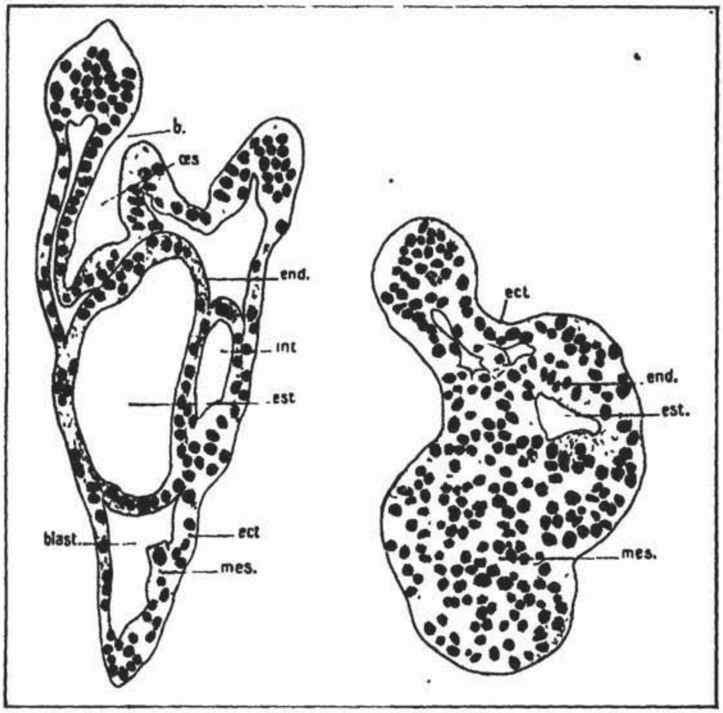

Figure 50. Cross section through sea urchin larvae.
Left: the normal larva. -- Right: larva exposed to the radiation of *Bact. tumefaciens*.
b = mouth; *blast* = blastophore; *ect* = ectoderm; *end* = endoderm; *est* = stomach; *int* = intestine; *mes* = mesenchyme; *oes* = oesophagus.

The most remarkable fact is perhaps the observation that the irradiation of the sperm alone or of the unfertilized ovum alone will suffice to bring about the same morphological changes. Thus, the radiation received by the single egg or sperm cell affects the entire future development of the larva, and the stimulus received by such a cell affects only one part of the offspring, i. e. the mesenchymatic cells.

3. Metamorphosis of Amphibia. The role of mitogenetic radiation in the metamorphosis of amphibia has been studied extensively by Blacher and his associates. The most detailed investigation concerned the development of tadpoles.

The following developmental stages are distinguished:

Stage I : hind legs differentiated
II : hind legs scarcely motile
IIIa: hind legs actively motile, belly rounded, forelegs not visible through the skin.
IIIb: belly becomes lean, elbows of forelegs distend the skin.
IV : forelegs are out; tail has full length
Va: about one-fourth of the tail is resorbed
Vb: about one-half of the tail is resorbed
Vc: about three-fourths of the tail is resorbed
VI : the entire tail is resorbed; metamorphosis completed.

Radiation arises from the resorbed tissues. The newly-formed fore or hind legs do not radiate. The intensity of radiation was estimated by the amount of the mitogenetic effect (yeast bud method) which is not a particularly good measure. The results are shown in fig. 51. All organs radiate only during their resorption. The gills initiate the process, followed closely by the intestine which is shortened considerably, the main shortening occurring at the time the forelegs develop. When the gills are almost completely resorbed, the tail begins. Other parts of the tadpole were tried, e. g. the back of the skin, but they did not radiate.

In a later paper, Blacher and Liosner (1932) estimated the intensity of blood radiation of *Rana ridibunda* during metamorphosis. They ascertained the minimal time necessary to bring about a distinct mitogenetic effect. The results were

at Stage II : 5 minutes; relative intensity: normal
IIIa: 30 minutes — one-sixth normal
IIIb: 15 seconds — 20 times normal
IV : 5 minutes — normal
Vb: 30 seconds — 10 times normal
VI : 5 minutes — normal

The strongest blood radiation coincides with the stage where gills, intestine and tail are all near the maximum of radiation. Otherwise, there is no parallelism. The second pronounced maximum of blood radiation occurs after gills and intestine have completed their metamorphosis.

Blacher and associates (VII) further showed that there was a relation between the resorbed tissue and the developing limbs, and that transplantation of gills increased the growth rate of the corresponding foreleg. Removal of the gill, or the freeing of the foreleg from the gill cavity where resorption of the gills occurs, retarded growth. Finally, it could be shown that a tadpole lying in a quartz-bottomed vessel will show a more rapid growth of the legs if the bottom is placed over pulp of tissues in the process of resorption. The foreleg to be irradiated was freed from its cavity by a cut in the covering skin, while the other leg which served as control remained covered, and therefore shaded against radiation. The result of the operation as such was that in the vessels with glass bottoms, the freed leg grew a little more slowly than its mate in the cavity: 6.5% ± 0.8 less in one series of 22 tadpoles, and 4.5% ± 1.3 less in a second series with 44 tadpoles. In the quartz-bottomed vessels, however, the freed legs grew more rapidly; 4.1% ± 1.1 more in one series of 33 tadpoles and 7.4% ± 1.2 in the other series with 51 tadpoles. The actual

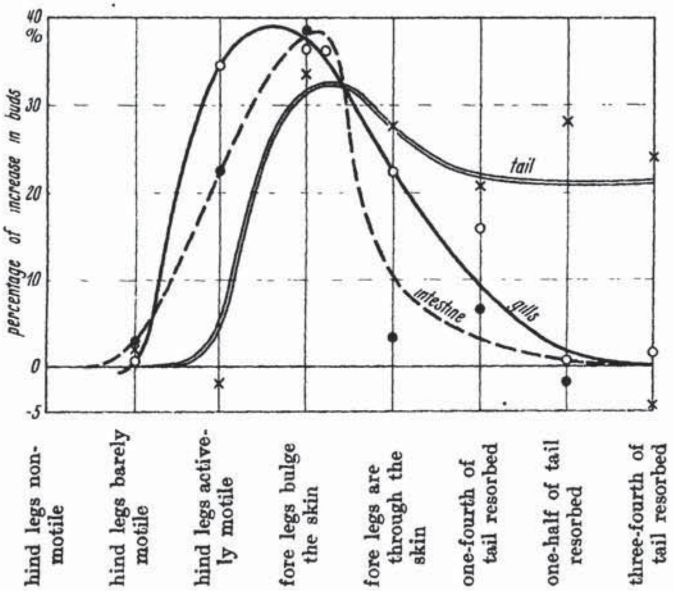

Figure 51. Intensity of radiation of the resorbed organs of frog tadpoles, during successive stages of metamorphosis.

gain by irradiation was therefore 10.6% ± 1.4 in the one set and 12.1% ± 1.8 in the other set of experiments. The gain is 7 times that of the average error.

The triton *Pleurodeles Waltii* shows similar radiations (Blacher etc., III), though less intense; and the axolotl *Ambystoma tigrinum* which retains gills as well as tail throughout its life radiates still less.

Experiments were made also with *Drosophila* larvae (Blacher etc., V). They do not radiate until about ready to pupate. The maximum intensity is reached 24 hours after pupation, and radiation nearly ceases after 30 hours.

There is no direct proof that in these cases, the radiation of the resorbed parts had any distinct form-giving effect. Positively established is only the radiation of the resorbed tissue, and an accelerating effect of this radiation upon the *rate* of development of the organs concerned.

4. Change of Chromosome Number. It has been repeatedly observed that polyploidy occurs fairly frequently in some individual branches of tomato plants infected with *Bacterium tumefaciens*. Kostoff and Kendall (1932) conceived the idea that polyploidy might be somehow linked with the strong mitogenetic radiation which is known to be emitted by this bacterium.

To test this, 120 tomato plants were inoculated with *Bact. tumefaciens*. After the development of the tumor, the plants were cut off 3 to 5 cm. above the tumor in order to induce the development of new shoots. Seven shoots arose directly in the tumor and of these, 5 could be rooted. One of them proved to be tetraploid.

While these data are, of course, too scant to permit the drawing of conclusions, they are suggestive of a new cause for polyploidy.

5. Parthenogenesis by Radiation. Reiter and Gabor after having verified Gurwitsch's statement that there is always radiation around a wound, refer to the theory of Bataillon that the developmental mechanism of an egg is released by the wound produced through the entrance of the spermatozoon. He had succeeded in causing cell division in unfertilized frogs' eggs by very fine needle pricks; only a very few percent of the eggs thus treated initiated development, but in some cases, Bataillon obtained tadpoles and even frogs by this method.

Reiter and **Gabor** speculated that this effect was in all probability due to the mitogenetic radiation of the wound (see wound radiation p. **173**), and they reasoned that in this event, radiation alone, without wounding, should produce the same result. They exposed unfertilized eggs of salamanders and frogs, obtained with great precautions from the females, to various sources of ultraviolet. Of all the wave lengths tried, only those of 3340 Å proved efficient (Attention may be called again to the circumstance that **Reiter** and **Gabor** have always claimed that only the rays around 3340 Å are mitogenetic, see p. **60**). Even with this wave length, only 2 unfertilized eggs of the salamander could be induced to develop, the first division being completed 3 hours after the 5-minute period of irradiation. A repetition caused only one such egg to develop. In the controls, as well as in all experiments with longer or shorter irradiation, or with different wave lengths, the eggs died in a short time. The number of eggs exposed is not given, except that it was stated that each group contained about 5 eggs. Of these "induced eggs", one developed to 32 cells, one to the 8-cell stage.

In a later experiment with "a few hundred" unfertilized frogs' eggs, 3 could be induced to start cell division by irradiation with monochromatic light of 3340 Å.

H. SYMBIOSIS, PARASITISM AND ANTAGONISM

It seems very likely that biological radiations play an important role in the mutual relationship not only of cells, but also of cell groups and multicellular organisms. This field of mitogenetic radiation has as yet scarcely been entered. A few isolated facts stand out plainly.

From the material mentioned in the preceding chapters, biological radiations appear to be largely stimulating, or beneficial. Possibly, this conception is wrong. It may be that only for reasons of technique, we have been able to observe for the most part this one type.

It has been shown that as a rule, unicellular organisms further each other's growth. The radiation from yeast stimulates the cell division of many bacteria, and the rate of germination of mold spores. Similar effects with multicellular organisms are imagineable, but not proved. The radiation from an onion root will stimulate cell division in another root under laboratory conditions, but that can hardly occur under normal conditions of plant growth, with the soil absorbing all radiation.

The role of radiation in parasitism is strongly suggested, but not proved in the case of plant tumors caused by *Bact. tumefaciens.* MAGROU (1927*c*) observed that cell division in the tumor was not limited to the immediate presence of the bacterial cells. However, it was not attempted to prove that a tumor would be formed even if chemical effects by bacteria were excluded from action through a wall of quartz.

The abnormal proliferations on the leaves and stems of plants due to insect eggs (commonly called galls) are usually attributed to chemical irritation by the larva, but it does not seem impossible that the young larvae radiate strongly during their early period of growth, and cause abnormal proliferation of tissue by irritation through ultraviolet rays.

Parasitic existence is limited to a very few species of plants or animals. It is customary to assume that for chemical reasons, all other plants and animals are prevented from living as parasites. The assumption that specific radiations also play a part offers itself naturally. However, this assumption would imply very specific radiations: either beneficial, which favor one species, but none of its nearest relatives, or, more probably, antagonistic radiations which prevent growth, or cell division, of all species except the parasite; for some reason, the parasite is not affected by the harmful radiation. The assumption of specific radiations has very little experimental evidence for its support. While we are still far from knowing all sources of mitogenetic rays, all well-known radiations arise from entirely unspecific biochemical processes such as proteolysis, glycolysis, oxidation, etc., processes which are common to most plants and animals. The pathogenicity of the typhoid bacterium to man, but not to rats could not be explained by different radiations from the two hosts, at our present state of knowledge. But it must be admitted that we have no chemical explanation either.

On the other hand, there has never been observed any specific action of definite wave lengths upon definite organisms. All experiments so far point to the conclusion that any radiation between 1900 and 2600 Å can stimulate the division of any cell, plant or animal, unicellular or part of a tissue, providing that the physiological conditions permit (see e. g. fig. 28 p. 49).

It is known, of course, that an overdose of rays will not stimulate, and might even retard growth, but there is no evidence that parasites are more tolerant to radiations of this kind than related, non-parasitic species. While the assumption of specific radiations would be a very convenient explanation for some problems in parasitism, it has as yet no experimental foundation.

Antagonistic Radiations: The only good case of specific antagonistic radiations is the investigation of Acs (1933) who experimented with microorganisms known to antagonize each other when grown simultaneously in the same culture, such as *Bacillus anthracis* and *Pseudomonas pyocyanea*, or yeast with staphylococci or streptococci.

Acs used for his experiments cultures which were 6—8 hours old, i. e. at the stage of rapid growth, and exposed the one pure culture to the radiation from the other. In this way, he obtained distinct retardation of growth. Irradiation of *B. anthracis* for 1 to 2 hours by *Ps. pyocyanea* gave growth retardations of 22 to 136%, in 14 experiments. In two experiments, he reversed the arrangement, using *B. anthracis* as sender, and found *Ps. pyocyanea* 42 and 48% retarded. The same organism was used simultaneously to irradiate *Bacillus ratimors*, which was not retarded, but stimulated 42 and 56%. Yeast radiation was found to retard staphylococci very distinctly, in 5 experiments, and also streptococci in the one experiment made.

Since such selectivity of antagonistic radiation cannot be explained by our present knowledge, a more detailed spectroscopic investigation of such types might add greatly to our understanding of the significance of biological radiations.

Different from the specific harmful radiation which injures one species, but stimulates other species, are the generally harmful human radiations observed by CHRISTIANSEN and by BARNES and RAHN (see p. 184) which are linked with certain pathological conditions. The radiation may be truly specific; only one species of yeast, *Saccharomyces Mycoderma punctisporus*, could be killed, while other yeast species were retarded, or not at all affected.

The example of the sea urchin larvae (p. 86 and 164) is quite characteristic of a harmful effect produced by a primarily beneficial radiation. In this case, the harm is probably done by an overdose and not by any specific wave lengths since such widely different sources as chemical oxidation, glycolysis by yeasts or streptococci, and proteolysis by staphylococci gave the same results. The main cause of the harmful effect in this case is the difference in sensitivity or reactivity, the mesenchyme cells being the only ones which were stimulated out of proportion.

I. THE HEALING OF WOUNDS

When wounds begin to heal, this is usually accomplished by mitosis of the cells nearest to the wound. In most cases, as with grown animals or plants, this would mean that cells which have already come to a resting stage, revert to multiplying cells. This resembles somewhat the situation of old yeast or bacterial cells transferred to a fresh medium. Something like a rejuvenation process of the old resting cells is necessary before they can divide again. It was precisely at this stage that we found mitogenetic radiation to be most effective upon bacteria or yeasts.

The first discussion concerning the possible rôle of biological radiation in the healing of wounds occurred in 1929. **Haberland** had found that full-grown, but young leaves of *Sedum spectabile* and *Escheveria secunda* can be torn without rupturing the cells of the mesophyll. They separate, and leave a dry surface. Such a surface will not "heal", i. e. the cells show no signs of division if the torn leaf is kept in a moist chamber. They will begin to divide, however, if smeared with juice of crushed leaves. They will also reproduce if the leaf is cut instead of carefully torn, leaving the wound surface wet with cell debris.

Gurwitsch (1929) expressed his opinion that such new division of old cells could not take place without a mitogenetic stimulus, and that very probably it came from the cell pulp. **Haberland** (1929) attempted to discover whether radiation by sedum leaf pulp would cause a healing or renewed mitosis of a dry, torn leaf. He found that leaf pulp did *not* induce any division of cells when held at short distances from the injured leaf and concluded that the wound hormones act chemically and not physically.

Gurwitsch (1929) found, using yeast as detector, that pulp of sedum leaves did not radiate when fresh, but would do so after 18 to 24 hours, losing this power again after 48 hours. He states that in his opinion, radiation alone will not cause the healing process in this case; that there are chemical requirements besides the physical ones; but, that radiation is *one* of the necessary factors.

While the argument concerning the sedum leaf wounds has never been settled experimentally, Bromley (1930) has proved that the tail of the salamander *Ambystoma tigrinum* radiated strongly after being amputated. It is not the new tissue which radiates, but the old cells immediately under the newly-formed tissue. Exposing yeast for 18 minutes to the ground tissue, the following "induction effects" were obtained:

1	2	3	5	6	12	24	hours after amputation
6.2%	2.1%	29.6%	41.1%	39.9%	48.4%	56.4%	induction

It requires 3 hours for radiation to be established. The radiation of the injured tissue reaches a maximum after 1—2 days, decreases decidely and reaches a second maximum after 5 –6 days. This agrees with the two maxima in pH of healing wounds as observed by Okuneff (1928).

The same double maximum could be observed with wounded tadpoles of the frog *Pelobates fuscus*, by Blacher, Irichimowitsch, Liosner and Woronzowa (1932b). They cut 10 mm. from the tip of the tail, and after different times, removed the old tissue which produced the regeneration, ground it up and used the pulp to irradiate yeast plates. The threshold time necessary to produce distinct mitogenetic effects was the measure of intensity. In Table 50, two distinct maxima are seen at 12 and 36 hours after wounding, and a minimum at 24 hours, and a great drop in intensity after 4 days.

Very interesting was the observation by the same authors that the blood of the tadpoles changed its radiation decidedly upon wounding of the tail. The blood of normal, uninjured tadpoles produced a marked effect in 5 minutes. 12 hours after injury, 1 minute of exposure to blood sufficed for a similar effect; 24 hours after injury, 2 minutes were required, and on the fourth day, an exposure of 15 minutes became necessary. This means that at this stage, the blood radiated less than half as strongly as that of uninjured tadpoles, and it stayed at this low level during the entire 18 days of the experiment.

By this long-continued effect of the wound upon blood radiation, the entire body is affected and brought into play.

Table 50. **Induction Effects Produced in Yeast by Irradiation with Old Tissue Bordering the Regeneration of Wounded Tadpole Tails** (*Pelobates fuscus*)

Time after Wounding	Irradiated for							
	10 sec.	15 sec.	30 sec.	1 min.	2 min.	5 min.	10 min.	15 min.
6 hours	—	—	0	**+28**	—	—	—	—
12 ,,	+2	+18	**+29**	+ 7	—28	—29	+ 7	—
24 ,,	—	—	—	— 5	**+18**	—	—	—
36 ,,	—	+ 4	**+36**	—	—	—	—	—
2 days	—	+ 2	**+26**	+12	—16	— 9	—23	—
3 ,,	—	—	+ 3	**+24**	—	—	—	—
4 ,,	—	—	—	—	—	—	— 8	**+36**
5 ,,	—	—	—	—	—	—	+ 1	**+52**
6 ,,	—	—	—	—	—	—	+ 2	**+28**
8 ,,	—	—	—	—	—	—	+ 2	**+21**
11 ,,	—	—	—	—	—	—	**+25**	—
12 ,,	—	—	—	—	—	—10	**+20**	—
18 ,,	—	—	—	—	—	+ 2	**+31**	—

Samarajeff (1932) repeated the tadpole and salamander experiments with earthworms. He cut them through, leaving them in earth, and found that the first mitogenetic effect could not be detected until 20 hours after inflicting the wound. Radiation was weak throughout, but was noticeable even after 6 days.

Considering that regeneration in wounds is accompanied by a radiation of the injured tissue, it seemed probable that this radiation was essential in bringing about healing. Blacher, Irichimowitsch, Liosner and Woronzewa (1932a) succeeded in proving this by producing a more rapid regeneration of wounds by biological irradiation. More than 500 tadpoles were used for this proof. Into their tails were cut triangular wounds, as in fig. 52. The rate of healing was measured as shown in the same figure, under a low power lens, and it is here recorded in μ of new

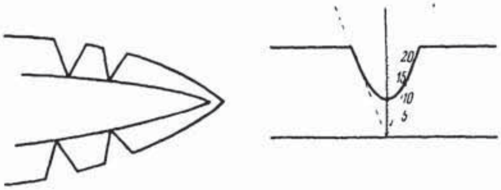

Figure 52.
At left, method of wounding the tadpole tails for irradiation from below; at right, method of measuring the amount of regeneration.

Table 51. Amount of Regeneration in the Wounds of Tadpole Tails, Irradiated from Below

Irradiated during	Irradiated				Controls			
	No. of tad-poles	new growth, in μ under side	new growth, in μ upper side	Induction Effect	No. of tad-poles	new growth, in μ under side	new growth, in μ upper side	Induction Effect
1st 24 hours	18	582	513	+13	16	444	480	—7
after	10	324	210	+54	17	246	243	+1
wounding	16	645	555	+16	12	477	528	—10
2nd 24 hours	20	777	678	+14	20	744	753	—1
after	17	576	459	+26	16	489	498	—2
wounding	16	666	528	+26	14	657	705	—7
3rd 24 hours	14	849	765	+11	14	786	762	+3
after	16	585	486	+20	18	561	558	+1
wounding	19	1014	813	+25	16	702	645	+9

growth from the deepest part of the wound. The tadpoles were placed in dishes with quartz bottoms which rested on a pulp of tadpole tails and intestine. This pulp is known to radiate strongly, and the wounds on the under side of the tail were thus exposed through quartz, while the wounds on the upper side were not. The controls were in similar dishes with glass bottoms. These experiments, of which a few are reproduced in Table 51, showed that in the controls, there was no difference in the rate of regeneration between the upper and the under side of the tail. Irradiation produced in all series of experiments a distinctly more rapid healing of the irradiated wounds.

However, this statement needs some modification as a careful checking of the individual wounds showed. When all the upper (non-irradiated) wounds of the exposed tadpoles were compared with those of the controls, there was no difference when the tadpoles were exposed for 24 hours immediately after wounding. There was also no difference when the tadpoles were left without treatment for 48 hours and then exposed for 24 hours. If, however, the tadpoles were left untreated for 24 hours and then exposed for 24 hours, the upper wounds had healed 26% less than the controls. A comparison of the lower wounds showed an increase in healing of 33% when irradiated at once, but no difference when irradiation began after 24 hours.

This signifies that irradiation immediately after injury affects only the irradiated wounds, stimulating them; irradiation 24 hours after wounding does not stimulate these, but retards the others, manifesting an effect which is only apparently beneficial. Irradiation 48 hours after wounding again produces a true stimulation. There is a suggestion of the same double maximum which we have already encountered. No explanation has been attempted.

The proof that the rate of healing of wounds can be accelerated by mitogenetic rays, may be of importance for the future of wound treatment. Two treatments might be, partly at least, explained by mitogenetic rays. One is the beneficial influence of the presence of Lactobacilli. NORTH (1909) and GILTNER and HIMMELBERGER (1912) applied cultures of *Lactobacillus bulgaricus* to inflamed mucous membranes (gonorrhoea, hay fever, conjunctivitis, utero-vaginal affection of cows after abortion) and to suppurating wounds with very good healing results. The good success is ascribed to the lactic acid. The other method is the rapid healing of wounds infested with maggots of flies. While doubtless the present explanation is correct, that the continuous removal of pus and dead tissue cells by the maggots induces more rapid healing, it may well be that in addition, there is a mitogenetic effect upon regeneration by the larvae.

J. THE CANCER PROBLEM

The word *cancer* is not very accurately defined. It is meant to indicate the most common form of malignant tumor. From the physician's point of view all tumors are neoplasms. Since the authors do not feel competent to pass judgment upon medical definitions, they have adopted the following of FELDMAN's (1932):

"With certain reservations, a neoplasm may be defined as an autonomous proliferation of cells, non-inflammatory, which grow continuously and without restraint, the cells resembling those of the parent cell from which they derived, yet serving no useful function, and lacking orderly structural arrangement."

"Carcinoma is preferable to the older term cancer as designating tumors of epithelial origin that are malignant."

"Sarcoma refers to a tumor consisting of immature connective tissue elements, that is clinically or histologically malignant."

It has already been mentioned repeatedly in previous chapters that contrary to the very weak radiations of the normal tissues of grown animals, the malignant tumors radiate strongly, while the benign do not.

The first study on the relation between tumors and mitogenetic rays, however, was the investigation by the MAGROUS (1927a and b) of plant tumors caused by the crown gall organism, *Bacterium tumefaciens*. They proved, with onion roots as detectors, that cultures of the bacterium radiated; they also showed (1927c) that in the tumors, cell division had taken place quite removed from the location of the bacteria. This made a physical effect probable, but did not exclude chemical effects. They did not produce experimentally plant tumors by radiation.

Soon afterwards, the carcinoma problem in man and animals was attacked from various angles; (1) the radiation of cancerous tissues; (2) the radiation of blood of cancer patients, as a means of diagnosis; (3) the origin of cancer.

Radiation of Cancerous Tissues: The strong radiation of cancerous tissue as contrasted with the non-radiating normal tissue has been established in a number of cases by various investigators, e. g. GURWITSCH, REITER and GABOR, SIEBERT, STEMPELL, GESENIUS, and by various methods. It is one of the most definite facts of mitogenetic radiation. It is also an established fact that only malignant tumors radiate.

A. and L. GURWITSCH (1929) and KISLIAK-STATKEWITSCH (1929) had found that there were two kinds of carcinoma radiations; one requires glucose in order to emit rays, while the other occurs primarily in the necrotic parts of cancerous tissue which are not capable of glycolysis. By comparing the spectra of these two different types with the known spectra, it could be shown that the one was plainly a glycolytic radiation while the lines emitted by the necrotic parts of the tumors agreed with those of proteolysis (see Table 52). An analysis with strips of 50 to 60 Å was sufficient to prove the difference.

The nucleic acid spectrum is also found in carcinoma itself if the exposure is sufficiently long; the proper time for the production of a glycolytic spectrum is too short for that of nucleic

Table 52. **Analysis of the Two Spectra of Carcinoma**
(the numbers indicate induction effects)

Carcinoma Type	Wave Length in Å	in situ	in Ringer's solution + glucose	Glycolysis of Normal Blood
Intact Metastases	1900—1950	50	44	25
	1950—2010	55	33	32
	2010—2070	—3	—9	5
	2070—2150	1.3	3	—1
	2150—2220	50	66	30
	2220—2340	0	0	2

		necrotic carcinoma	serum albumin + pepsin
Necrotic Parts	1900—1960	—1.5	—3
	1960—2020	38	50
	2020—2080	53	80
	2080—2140	45	70
	2140—2210	8	1
	2210—2290	2.5	11.6
	2290—2390	44	50
	2390—2430	42	36

acid. This difference in intensity of various reactions occurring simultaneously adds greatly to the difficulties of spectral analysis.

Absence of Blood Radiation: The second outstanding and thoroughly established fact is the absence of blood radiation in cancer patients (see fig. 46, p. **153**). Gesenius (1932), in summarizing 3 years of clinical experience, states that patients with teratomes (monstrosities) and probably with mixed tumors never show blood radiation; while normal radiation is observed in all cases of sarcoma[1]), hypernephroma (benign kidney tumor), glioma (benign tumor of the brain, retina or auditory nerve), and myoma (benign muscle tumor), even in severe anemia caused by bleeding of the myoma.

Braunstein and Heyfetz (1933) determined the time when blood radiation decreases. They inoculated rats with tumor

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[1]) This seems strange since the sarcoma tissue shows the same strong radiation as carcinoma tissue.

Table 53. Glycolysis and Radiation in Blood from Rats Inoculated with Tumor Cells

	No. of Rats	Decrease of Sugar in Blood, in mg, after		Mitogenetic Induction
		1.5 hrs	3 hrs	
Normal Animals	14	50.1	67.6	+33.4
5 days after implantation of cancer	3	38	69	+26
6 days	6	40	66	+13
8 days	7	62	78	+ 7
10 days	7	43	70	+ 7
13 days	6	46	69	— 4
16 days	3	52	77	+ 5

cells through an incision of the skin of the back; after 6 days, a tumor the size of a barleycorn had developed. In short intervals, they determined the decrease in blood sugar, and the mitogenetic radiation (Table 53). While the latter decreased distinctly from the fifth day, glycolysis and sugar content were not affected. There is really much more to this negative induction effect of cancer blood than merely the absence of radiation. Heinemann (1932) very definitely found growth retardation when he used the actual rate of cell increase as a measure (Table 22, p. 73 and fig. 45, p. 153). As early as 1929, Lydia Gurwitsch and Salkind observed that the blood of cancer patients suppressed the radiation of normal blood upon mixing the two. Rats, injected with cell-free tumor extract, showed no blood radiation after 2, 3, and 4 days, but were normal again after 9 days. Injection of cytagenin (see fig. 45) caused a temporary increase of blood radiation. When cytagenin is discontinued, emission of rays drops to its negative level in 1 to 3 days.

Cancer Diagnosis: Blood radiation is so conspicuous even with patients suffering from many kinds of severe illnesses that its absence in cancer has been considered since the earliest observations as a diagnostic means. Gesenius reports (1932) that in pernicious anemia, radiation begins again after liver diet, while in carcinoma, even the complete removal of the tumor and radium treatment will not restore normal blood radiation.

Recently, KLENITZKY (1934) observed the return of blood radiation when every last trace of cancerous tissue had been removed.

Since practically all diseases producing decreased mitogenetic radiation in blood are readily recognized, cancer diagnosis by this method has been studied systematically. The best results obtained are very likely those recorded by HEINEMANN from one of the hospitals in Frankfurt, Germany. "On the one side, further observations with carcinoma suspects justified the conclusion that a positive mitogenetic effect excluded the possibility of a tumor with certainty. On the other hand, a negative effect changing promptly to positive after cytagenin injection, caused us repeatedly to search for a tumor and to prove it beyond doubt, though the preceding, most careful clinical investigation had given no reason to suspect tumors (e. g. a small carcinoma in the upper rectum which caused no direct pain)."

Very encouraging is further the summary by GESENIUS (1932) of 3 years experience in the Berlin University clinics. The method consisted in the decrease of yeast respiration by blood radiation (see p. 84). During the last year, only such cases in which diagnosis was uncertain were investigated.

Occasional exceptions have been observed; blood radiation has been found infrequently connected with severe carcinoma, and has rarely been absent in patients where autopsy revealed absence of carcinoma. Nevertheless, blood radiation is a valuable diagnostic means when used as a part of the clinical examination. Loss of radiation is independent of the size and location of the tumor. It remains absent after removal of the tumor, and cannot be used as a test for the success of treatment.

Origin of Cancer: It has been stated repeatedly that normal adult tissues radiate very weakly while normal blood radiates strongly; in cancer, the opposite takes place, the new growth radiating strongly while the blood ceases to do so. These facts plainly suggest that the growth-stimulating source of radiation is removed from the blood and concentrates in the neoplasm. However, this simple assumption does not agree with our explanation of these radiations as originating from biochemical reactions. It has just been shown that sugar content and the rate of glycolysis in the blood of cancer patients are not greatly altered. For this reason, this explanation of the origin of cancer is not usually stressed.

Somewhat different is PROTTI's conception (1934*b*) who assumes that a cellular disorder may arise when blood radiation becomes very low, eventually resulting in a neoplasm. He proved his point by injecting a cell suspension of adeno-carcinoma into two lots of 12 mice each; one lot was being fed normally, while the other was on a starving ration. For the first 15 days, the tumors grew more rapidly in the starved mice whose blood radiation had decreased, on the day of injection, from about 50 to about 10. It must be remembered that cancer is most frequent in old age when blood radiation has a tendency to become very low.

PROTTI observed further that a mixture of neoplasmic cells and yeast cells *in vitro* resulted in a destruction of the neoplasmic cells while cells from normal tissues were not influenced by yeast. The same happened when the neoplasmic cells were separated from the yeast cells by a quartz plate. PROTTI calls this "cytophotolysis".

Repeated injections of yeast suspensions into the Galliera-Sarcoma of rats caused a liquefaction of the tumor, without pus formation, leaving finally a cavity with thin fibrous walls. Intravenous injections produced no effect upon the tumors. With the adeno-carcinoma of mice, injections of small amounts of yeast into the tumor stimulated its growth while large amounts retarded it. The same was the case with intravenous injections. Yeast heated to 60⁰ produced no effect which suggests, together with the abovementioned "cytophotolysis", that the results are not due to enzymes, but to radiation.

Cancers have been produced by frequent application of certain chemicals to the skin. Since the compounds found so far are not normal or pathological products of the human body, as far as is known, the discovery of their effect does not really explain the formation of cancers, but may give valuable suggestions towards the solution of the problem.

More important is perhaps the discovery that cholesterol metabolism is in some way connected with cancer. It has been claimed by SHAW, MACKENZIE, MORAVEK and others that cholesterol stimulates the growth of malignant tumors. ROFFO (1933) found the cholesterol content of the skin near cancerous or precancerous lesions to be much higher than the normal skin of the same patient. He found the frequency of skin cancer to be distributed in the following way, as average of 5000 cases:

skin of the face	95.51%
skin of the back of the hand	3.07%
scalp	1.02%
skin of the foot	0.50%

Kawaguchi (1930) had shown that the cholesterol content of the skin increases when it is exposed to sun light. According to Malcynski (1930), ultraviolet irradiation of healthy persons increases the blood cholesterol while with cancer patients, there is a decrease of 25 to 40%.

Roffo (1934a) studied the "heliotropism" of cholesterol very thoroughly. He proved it to take place only in living organisms. With white rats, sunlight as well as ultraviolet light increased the cholesterol content of the exposed parts of the skin. The wavelength of the ultraviolet was above 2300 Å. X-rays or radium rays produced the opposite effect.

Then, Roffo (1934b) studied the tendency for cancer developpment in white rats. 700 rats were kept in sunlight daily for not more than 6 hours, avoiding the hottest sun. After 10 to 11 months, 52% of all rats had developped cancer, which was exclusively on the naked parts of the body (ears and eyes mainly, also twice on the hind feet and once on the nose). 150 rats were exposed to ultraviolet light, of an intensity of 14 erythemal units, beginning with 5 minutes per day and gradually increasing the exposure to 6 hours. Within 4 months, every one of these rats showed tumors, and many of them had several tumors. Of the control rats receiving light from a tungsten filament lamp, not a single animal had developped a tumor. This agrees very well with the above-mentioned frequency of skin cancer on different parts of the human body.

The mode of development and the histology of the rat cancers corresponded exactly with that of human cancers.

The author has not been able to ascertain whether Roffo has tested the mitogenetic range of rays by itself, i. e. the range between 1800 and 2600 Å. It would be a splendid agreement if this range which is known to stimulate cell division could also be shown to produce abnormal cell division of the epithelial cells. The fact that sunlight can do this speaks very much against it since sunlight contains no wavelengths shorter than 2700 Å. However, very small amounts, such as have been shown to produce mitogenetic effects, might possibly be present (at least in Buenos Aires where these experiments were made). The much stronger effect of artificial ultraviolet containing also the shorter waves suggests that the shorter waves are more efficient in bringing about abnormal cell division.

Attention may be called here to the effect of radiation of oxy-cholesterol upon yeast cells. It is impossible to say whether any relation might exist between oxy-cholesterol and the cancer problem.

K. RADIATIONS OTHER THAN MITOGENETIC

In Chapter IV, three types of radiation from organisms have been mentioned which are not identical with mitogenetic radiation. One was entirely different, and does not really belong in this group, namely the *Beta*-radiation of the potassium in the cells. Then, there was *necrobiotic* radiation. This type, though apparently stronger than mitogenetic rays, is emitted by dying cells, and does not have any definite biological purpose or effect as far as has been ascertained. It is an emission of energy typical for the chemical changes connected with death, but it is absorbed by the surrounding medium, and should it produce biological effects, it would be purely accidental.

The third group are the harmful human radiations, and they produce distinct biological effects. Quite commonly known are the reactions brought about by menstruating women: flowers wilt readily in their hands; menstruating women are excluded from collecting flowers for the perfume factories of France; mushroom beds are said to be utterly ruined by their mere presence; pure cultures of yeasts and bacteria become abnormal when handled by them (see p. 95), and the common belief that bread dough will not rise normally, and that pickles and sauerkraut packed by them will not keep, does not sound improbable, since Christiansen proved that wine fermentation was distinctly affected (see p. 96).

The facts as such can be considered fairly definitely established. Details may be found in the paper by Macht and Lubin (1923—1924). But it cannot be considered proved that the effect is of physical origin. The more common conception among medical men is that it is chemical, produced by a compound called menotoxin.

The intensity of this effect varies greatly with the individual. We have but rarely been able to obtain good results with wilting flowers. CHRISTIANSEN found the effect to be much stronger in summer than in winter.

Attention should be called here to the observation by HILL (1933) that certain persons kill bacteria on agar plates when placing their fingers right upon the seeded plate. In this case, it has not been decided as yet whether the effect is physical or chemical.

Probably the same type of harmful radiation has been observed in a very few cases of illness (p. 97); at least, the yeast was killed by emanation from the finger tips, and even from the face, through a thick quartz plate excluding chemical influences.

The Source of Harmful Human Radiations: MACHT and LUBIN (1923—1924) came to the conclusion that the "menotoxin", i. e. the compound in the blood during menstruation responsible for the various phenomena just mentioned, must be either oxycholesterol or a closely related compound. This induced BARNES and RAHN (1933) to test whether this compound radiated. The results were quite definitely positive. Out of 5 batches of oxycholesterol made at different times, 4 killed Mycoderma during the first day when exposed continuously through quartz, but not on the second day. With Batch No. III which had no effect upon the yeast, an error was made in the separation of the oxycholesterol from the other reagents; on account of this, the exposure could not be started until one day later, and by this time, this product usually has lost its power. The colorimetric tests mentioned for oxycholesterol can be obtained long after radiation has become too weak to be detected biologically.

Since it is not probable that a compound as such radiates, it appears more likely that the radiation comes from some reaction of the cholesterol or oxycholesterol, and we would think above all of oxidations. In the above experiments the oxycholesterol was emulsified with a little water to make oxidation possible.

Why this radiation is harmful instead of stimulating, cannot be stated at present. The simplest assumption would be that the death of the Mycoderma cells is due to over-exposure because in

Table 53a. Effect of Oxy-cholesterol upon Mycoderma through fused Quartz

time after manufacture of oxy-cholesterol	Batch I	Batch II	Batch III	Batch IV	Batch V
1 day	dead	dead	normal (stimulated)	mostly dead	dead
2 days	—	—	—	normal	retarded
3 days	—	—	—	stimulated	—
4 days	greatly retarded	greatly retarded	—	—	—
5 days	slightly retarded	—	—	—	normal
6 days	normal	—	—	—	—
7 days	normal	—	normal	—	—
8 days	normal	—	—	—	—
14 days	—	—	normal	—	—

all experiments the yeast was exposed continuously to this radiation. However, complete killing of all cells is not usually observed even in over-exposure, and only rarely was stimulation found. The other alternative that we are dealing with different wave lengths does not seem very probable either. All wave lengths between about 1800 and 2600 Å give the same mitogenetic effect (see fig. p. 49), and no exceptions are known; the oxycholesterol radiation passes through quartz but not through glass, and can therefore be hardly anything but ultraviolet. An analysis of the spectrum may give the explanation.

Oxycholesterol in the Body: A study of the distribution of cholesterol and oxycholesterol in the fats of the human skin, by Unna and Golodetz (1909) shows that oxycholesterol is not present in the normal cellular fats but only in secreted fats (Table 54). An exception is the fat in the finger nails and toe nails. This agrees quite well with our observation that radiation was obtained from hands (and feet) and from the face where sebacious glands exist, but not from the chest where they are very rare.

While a good deal of attention has been paid in recent years to cholesterol metabolism, very little is known about oxycholesterol in the body. Even its chemical composition is not certain. The only more recent investigation on oxycholesterol in the body is that by Pfeiffer (1931). According to his analyses, the largest

Table 54. **The Cholesterol and Oxycholesterol Content of Human Skin Fat**

		melting point °	Unsaponifiable fraction %	In % of the unsaponifiable fraction		
				free Cholesterol	Cholesterol Esters	Oxycholesterol
secreted fats	Comedon fat (blackhead)	53.0	30.50	9.16	--	very much
	hand sweat . . .	46.5	28.40	5.40	0.79	very little
	foot sweat	36.5	22.30	18.70	1.60	fairly much
cellular fats	surface skin fat .	48.0	32.20	50.00	2.06	0
	horny skin fat . .	51.0	36.30	54.00	8.91	0
	nail fat	38.0	41.60	43.70	—	fairly much
	vernix caseosa . .	38.0	36.00	45.00	8.38	0
	subcutis	liquid	1.15	15.80	0.04	0

amount of oxycholesterol, on the basis of total solids, is found in the brain (0.44%), next are bone marrow, adrenals and bile (about 0.29%), followed by the liver (0.09%). Lowest are the erythrocytes with 0.003%. While previous authors, e. g. UNNA and GOLODETZ, estimated the oxycholesterol colorimetrically, PFEIFFER determined it by the difference in melting points between cholesterol and oxycholesterol. The results may therefore not be comparable, on account of the uncertain chemical composition. It has been stated already that oxycholesterol radiation ceased when the colorimetric tests were still very strong. The relation between this substance in the tissues and radiation is therefore not definitely established.

In the preceding chapter, attention has been called to the close relation between cholesterol and cancer. The above results suggest that an investigation about the role of oxy-cholesterol in connection with the cholesterol metabolism in cancer might yield interesting results.

OUTLOOK

The biological part of this book has been written by a biologist who is convinced, from his own experiences as well as from the study of literature, that mitogenetic radiation exists. He has realized that it is difficult to prove it because we are dealing with an extremely weak effect, and with very sensitive detectors. Above all, we are dealing with an entirely new phenomenon, and consequently cannot predict which changes of technique might increase or decrease the effect.

It does not speak well for the present status of science that it has not been possible to settle definitely, in the course of 12 years, the question of the existence of this radiation. The fault lies equally with the two groups of contestants, those for and those against radiation.

The facts are these: GURWITSCH and a number of his pupils and also many other investigators have presented a very large amount of experimental data to show that mitogenetic radiation exists. Many others have repeated these experiments, following directions as exactly as they were given, and obtained no mitogenetic effect. Several of the latter group have claimed therefore that they have disproved biological radiation. Such claim is unscientific as has already been pointed out in the foreword. The only way to disprove any theory is to obtain the same results, and to show that they are due to another cause. Some such attempts have been made (e. g. MOISSEJEWA, LORENZ) but they have not been carried far enough, or have been contradicted by other, more recent investigations. Most of the critics dismiss the question with the simple statement that all so-called mitogenetic effects are within the limits of experimental error.

Let us realize from the beginning that the differences of opinion center around two essentially different points; one is the existence of the biological effect, and the other is its interpretation as an ultraviolet radiation. A different interpretation will not make the effect less important for biology. The effect is the important thing; the explanation is secondary. After all, only the facts remain permanent in science, while the theories come and go.

The difficulties in deciding the existence of the mitogenetic effect are to be sought largely in the sensitivity of the methods. It is evident that this point can be settled only by biological experiments. Physical measurements can tell only whether or not it is caused by radiation, but the absence of radiation does not disprove the biological effect.

Biological measurements are not at all simple. When higher organisms are used as detectors, the controls are not perfect. Several authors have questioned the use of one side of the onion root as control for the other side. It may be that both are affected. This objection may also be made to other tissues, e. g. the cornea. With unicellular organisms, where large numbers are used, the controls are as reliable as they can possibly be in any biological experiment. While it is easy to make yeasts or bacteria grow in the customary culture media, it requires a thorough understanding of their physiology to interprete differences of growth rate, and errors have been made in this respect by those opposing biological radiation as well as by those convinced of it.

The error in biological experiments is not as absolutely fixed as in physical or chemical methods, e. g. in an analysis where it can be stated reliably for all laboratories that the accuracy of the method is 0.005 g. In biology, it depends very much upon the choice of the organism (e. g. the variety of onion), the uniformity of the material, the treatment of the organisms before the beginning of the experiment, the uniformity of environment before and during the experiment, the ability of the experimentor to recognize and avoid disturbing or secondary influences. A fine example is the painstaking work of M. PAUL (1933) with onion roots.

As a result of the various factors creeping in, the error of the same method may be widely different in different laboratories or even in the same laboratory with different investigators. This explains the difference of error in the onion root experiments which was 10% with some investigators and 50% with others (see p. 58). When it comes to such delicate instruments as the GEIGER counter, even physical measurements show great differences (see Table 30a p. 92).

The frequently made statement that the biological investigators do not state the error of their methods is not in accordance with the facts. When the error or the reliability of the method is not stated as such, it is usually possible to compute the error from those experiments where no mitogenetic effects were obtained. This has been done by SCHWEMMLE for all earlier onion root experiments (p. 58) and the authors gave some similar calculations for the yeast bud method on p. 71. TUTHILL and RAHN have also published two sets of counts of yeast buds from many different parts of the same culture. GURWITSCH and his associates have stated repeatedly that with the yeast bud method, they consider any increase less than 15% over the control to be experimental error. The reliability of the yeast measurement by volume can be estimated from the data by BILLIG, KANNEGIESSER and SOLOWJEFF. HEINEMANN has stated that in his method of counting yeast with the hemacytometer, the mitogenetic effects were more than 3 times the experimental error. WOLFF and RAS have frequently given all individual counts of bacteria for one experiment (p. 78). Other error limits can be found on pp. 83 (JULIUS), 34 (BARTH) and 168 (BLACHER).

It is somewhat surprising to find that in critical summaries, some weak papers are quoted extensively while some of the best proof in favor of mitogenetic radiation is completely omitted. NAKAIDZUMI and SCHREIBER, working with the yeast bud method, omit the work of SIEBERT who published more detailed experiments than any other investigator in this field. KREUCHEN and BATEMAN also mention neither SIEBERT's papers nor the extensive work of WOLFF and RAS with bacteria which is the best material with this detector. It is only natural that any new development in science will attract speculative minds who generalize from a few experiments and come to conclusions which are not shared by the more conservative workers in this field. Any serious criticism should start with the most reliable and best founded papers.

On the other hand, the critics have good reason to disregard papers which give no precise account of methods or results. Probably the main reason why mitogenetic effects are still doubted has been the recording of results by merely giving the "Induction Effect" without mention of the experimental data from which the effect was computed. The actual number of mitoses in a cornea, the percentage of buds, or the yeast volume measured, tell a good deal about the performance of the experiment. Even the chemist whose methods are really standardized publishes not merely the formula of his new compound, but also the actual analytical data. Since the error in biological experiments varies with the investigator, the publication of the complete records would give the reader a conception of the reliability of any observed effects, even when the standard deviation has not been computed.

Another justifiable argument against the weight of some published papers is the lack of precision in the description of the method. Since the biological detectors do not respond at all times to mitogenetic rays, but only in a definite physiological state, it is of greatest importance to describe all details. Such statements as "8—10 hours at room temperature" are too indefinite; the term "yeast" means very little, and even such terms as "onion root" or "cornea of a frog" should be specified in much more detail since there are many different kinds of onions or frogs, and the roots as well as the number of mitoses in the cornea are affected by the season.

As an example may be mentioned the paper by SALKIND and PONOMAREWA (1934) which might be very important for the physiological explanation of the mitogenetic effect. However, the authors do not mention the age of the yeast culture, nor the medium on which it was grown; they give only the induction effect so that the reader does not know whether the controls had 3% or 25% of buds which would have given a suggestion at least of the condition of the yeast. Therefore, the value of the paper is largely lost.

It might be argued that the burden of the proof lies with the opponents since the mitogeneticists claim to have proved their point, but such an attitude would not be very helpful in solving the real problem. We are dealing with a very complex phenomenon, and both sides should do all they can to bring about a real understanding of the facts. The complexity is greatly increased by the occasional failure of the phenomenon for unknown reasons (p. 93). Errors have been made by the defendants of both sides of the argument, and the authors hope sincerely that by pointing out the mistakes and misunderstandings, a settlement of the question of mitogenetic radiation will be accomplished in a very short time.

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II. Blacher and N. W. Bromley, Mitogenet. Ausstrahlungen bei der Regeneration des Kaulquappenschwanzes. **121, 79**
III. — and O. G. Holzmann, Mitogenetische Ausstrahlungen während der Metamorphose der Urodelen. **123,** 230
IV. — and N. W. Bromley, Mitogenetische Ausstrahlungen bei der Schwanzregeneration der Urodelen. **123,** 240
V. O. G. Holzmann, Mitogenetische Ausstrahlungen während der Metamorphose bei *Drosophila melanogaster*. **123,** 266
VI. N. W. Bromley, Der Einfluß der primären Verheilung der Wunde auf die Entstehung mitogenetischer Ausstrahlungen in ihr. **123,** 274
VII. L. J. Blacher, M. A. Woronzowa, L. D. Liosner and W. N. Samarajew, Die mitogenetischen Ausstrahlungen als Stimulus des Wachstums der Vorderbeine bei der Metamorphose von *Rana temporaria*. **124,** 138

Blacher, L. J., and L. D. Liosner, 1932, Mitogenet. Blutstrahlung der Amphibien während der Metamorphose. Roux' Archiv **127,** 364 167
—, A. I. Irichimowitsch, L. D. Liosner and M. A. Woronzowa, 1932a, Einfluß der mitogenetischen Strahlen auf die Geschwindigkeit der Regeneration. Roux' Archiv **127, 339** 175
—, —, —, —, 1932b, Die mitogenetische Strahlung des Regenerats und des Bluts der Kaulquappen während der Regeneration. Roux' Archiv **127,** 353 174
— and W. N. Samarajew, 1930, Die Organisationszentren von *Hydra fusca* als Quelle mitogenetischer Ausstrahlungen. Biol. Zentr. **50,** 624 133
Böhmer, K., 1927, Menotoxin und Hefegärung. Deutsch. Z. f. d. ges. gerichtliche Medizin **10,** 448 95
Borodin, D., 1930, Energy emanation during cell division processes. Plant Physiol. **5,** 119 57
—, 1934, M-rays macro-effect and planimetric drop culture method. I. Internat. Congress of Electro-Radio-Biology, Venice 75
Braunstein, A. E. and P. A. Heyfetz, 1933, Die Glycolyse und die mitogenetische Strahlung des Bluts bei experimentellem Carcinoma. Biochem. Z. **259,** 175 179
— and A. Potozky, 1932, Oxydationsreaktionen als Quelle mitogenetischer Strahlung. Biochem. Z. **249,** 270 36
— and B. A. Severin, 1932, Der Zerfall der Kreatinphosphorsäure als mitogenetische Strahlungsquelle. Biochem. Z. **255,** 38 38
Bromley, N. W., 1930, Der Einfluß der primären Verheilung der Wunde auf die Entstehung mitogenetischer Ausstrahlungen in ihr. Roux' Archiv **123,** 274 174
— see also Blacher.

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Brooks see Nelson.

Brunetti, R., and C. Maxia, 1930, Sulla fotografia a la eccitatione delle radiazione del Gurwitsch. Atti Soc. fra il Cultori delle Science Med. e Nat. Cagliari **2** 90

Chariton, J., G. Frank and N. Kannegiesser, 1930, Über die Wellenlänge und Intensität mitogenetischer Strahlung. Naturwissenschaften **18**, 411 . . . . 50, 60

Christiansen, W., 1929, Das Monotoxinproblem und die mitogenetischen Strahlen. Ber. d. deutsch. bot. Ges. **47**, 357 86, 95, 161, 184, 185

Chrustschoff, G. K., 1930, Über die Ursachen des Gewebewachstums in vitro. I. Die Quellen der mitogenetischen Strahlen in Gewebekulturen. Archiv f. exper. Zellforschung **9**, 203 83

Coblentz, W. W., R. Stair and J. M. Hogue, 1931, A balanced thermocouple and filter method for ultraviolet radiometry, with practical applications. Bureau of Standards, Journ. of Research **7**, 723 48

Decker, G. E., 1934, Über die Schärfe mitogenetischer Spektrallinien. Archiv Sciences Biol. (Russ.) **35**, 145 37

Dietl see Polano.

du Bridge see Hughes.

Doljanski, L., 1932, Das Wachstum der Gewebskulturen in vitro und die Gurwitsch-Strahlung. Roux' Archiv **126**, 207. 83

van Doormal see Audubert.

Duggar, B. M. and A. Hollaender, 1934, Irradiation of plant viruses and of microorganisms with monochramatic light. Journ. Bact. **27**, 219 48

Emerson, R., and W. Arnold, 1932, A separation of the reactions in photosynthesis by means of intermittent light. Journ. Gen. Physiol. **15**, 391. 107

Feldman, W. H., 1932, Neoplasms of Domesticated Animals. Philadelphia, Saunders 177

Ferguson, A. J., 1932, Morphological changes in yeasts induced by biological radiation, Thesis, Cornell University 96, 161

— and Otto Rahn, 1933, Zum Nachweis mitogenetischer Strahlung durch beschleunigtes Wachstum der Bakterien. Archiv f. Mikrobiol. **4**, 574. . . . . 76, 78, 121, 126, 137

Frank, G., 1929, Das mitogenetische Reizminimum und -maximum und die Wellenlänge mitogenetischer Strahlen. Biol. Zentr. **49**, 129 . . . . 35, 39, 60

— and M. Kurepina, 1930, Die gegenseitige Beeinflussung der Seeigeleier als mitogenetischer Effekt betrachtet. Roux' Archiv **121**, 634 138

— and S. Rodionow, 1932, Physikalische Untersuchung mitogenetischer Strahlung der Muskeln und einiger Oxydationsmodelle. Biochem. Z. **249**, 321 . . . . 29, 34, 91, 109

Page

FRANK, G., and S. SALKIND, 1926, Die Quellen der mitogenetischen Strahlung im Pflanzenkeimling. Roux' Archiv **108**, 596 . . . 141

— and S. SALKIND, 1927, Mitogenetische Strahlung der Seeigeleier. Roux' Archiv **110**, 626 142

FRANK, M., 1921, Menotoxine in der Frauenmilch. Monatsschrift für Kinderheilkunde **21**, 474 95

FRICKE, HUGO, 1934, The chemical-physical foundation for the biological effects of x-rays. Cold Spring Harbor Symposia II, 241 . . 42

FRIEDRICH see SCHREIBER.

GABOR see REITER.

GATES see RIVERS.

GERLACH, W., 1933, Physikalisches zum Problem mitogenetischen Strahlung. Gesellsch. f. Morphol. u. Physiol. München **42**, 1 . 34

GESENIUS, H., 1930a, Über Stoffwechselwirkungen von GURWITSCH-Strahlen. Biochem. Z. **225**, 328 84

—, 1930b, Über die GURWITSCH-Strahlung menschlichen Bluts und ihre Bedeutung für die Carcinom-Diagnostik. Biochem. Z. **226**, 257 . . . . 85, 153

—, 1932, Blutstrahlung und Carcinom-Diagnostik. Radiobiologia **1** (2), 33 . . . . 85, 179, 180, 181

GILTNER, W., and L. R. HIMMELBERGER, 1912, The use of lactic acid cultures in combating infections of mucous membranes. J. Comp. Pathol. and Ther. **25**, 312 177

GOLISCHEWA, K. P., 1933, Die mitogenetische Spektralanalyse der Blutstrahlung am lebenden Tier. Biochem. Z. **260**, 52 . . 93, 150

GOLODETZ see UNNA.

GREY, J., and C. OUELLET, 1933, Apparent mitogenetic inactivity of active cells. Proc. Roy. Soc. B **114**, 4 92

GUILLERY, H., 1929, Über Bedingungen des Wachstums auf Grund von Untersuchungen an Gewebskulturen. Virchows Archiv **270**, 311 82

GURWITSCH, ALEXANDER, 1922, Über Ursachen der Zellteilung. Roux' Archiv **52**, 167 . . . . 59, 119

—, 1923, Die Natur des spezifischen Erregers der Zellteilung. Roux' Archiv **100**, 11 . . . . 54, 119

—, 1929a, Die mitogenetische Strahlung aus den Blättern von Sedum. Biol. Zentr. **49**, 449 . . . . 141, 173

—, 1929b, Über den derzeitigen Stand des Problems der mitogenetischen Strahlen. Protoplasma **6**, 449 . . . . 56, 60

—, 1932, Die mitogenetische Strahlung. Berlin, Springer. 380 pp. 63, 66, 70, 73, 74, 103, 107, 108, 113, 117, 127, 142, 147, 149

—, 1932a, Die mitogenetische Strahlung des markhaltigen Nerven. Pflügers Archiv **231**, 234. 155

—, 1934, Der gegenwärtige Stand des mitogenetischen Problems. I. Internat. Congress of Electro-Radio-Biology, Venice . . . 45, 146

Page

GURWITSCH, ALEANDER, — and L. GURWITSCH, 1925, Über den Ursprung der mitogenetischen Strahlen. Roux' Archiv **105**, 470 . 55

—, —, 1929, Die mitogenetische Strahlung des Carcinoms. II. Z. f. Krebsforschung **29**, 220 178

—, —, 1932a, Die mitogenetische Spektralanalyse: IV. Das mitogenetische Spektrum der Nukleinsäurespaltung. Biochem. Z. **246**, 124 38

—, —, 1932b, Die Fortleitung des mitogenetischen Effekts in Lösungen und die Beziehungen zwischen Fermentätigkeit und Strahlung. Biochem. Z. **246**, 127 . . . . 43, 109

—, —, 1934, L'analyse mitogenetique spectrale. Vol. IV of Exposés de Physiologie, Paris, Hermann and Cie., 39 pp. 40

GURWITSCH, ANNA, 1931, Die Fortpflanzung des mitogenetischen Erregungszustandes in den Zwiebelwurzeln. Roux' Archiv **124**, 357 111

—, 1932, Die mitogenetische Strahlung der optischen Bahn bei adäquater Erregung. Pflügers Archiv **231**, 255 . . . . . 157, 159

—, 1934a, L'excitation mitogénétique du système nerveux central. Ann. de Physiol. et Physicochem. biol. **10**, 1153 157

—, 1934b, L'excitation mitogénétique du système nerveux pendant l'éclairage monochromatique de l'oeil. Ann. de Physiol. et Physicochem. biol. **10**, 1166 158

GURWITSCH, LYDIA, 1931, Die mitogenetische Spektralanalyse: II. Die Spektren des Carcinoms und des Cornealepithels. Biochem. Z. **236**, 425 38

— and N. ANIKIN, 1928, Das Cornealepithel als Detektor und Sender mitogenetischer Strahlung. Roux' Archiv **113**, 731 84

— and S. SALKIND, 1929, Das mitogenetische Verhalten des Bluts Carcinomatöser. Biochem. Z. **211**, 362 . . . . 65, 152, 180

HABERLANDT, G., 1929, Über „mitogenetische Strahlung". Biol. Zentr. **49**, 226 173

HALL see JONES.

HAUSSER, K. W., and K. H. KREUCHEN, 1934, Quantenausbeuten bei Lichtzählern. Z. Techn. Physik **15**, 20 92

HARVEY see TAYLOR.

HEINEMANN, M., 1932, Cytagenin und mitogenetische Strahlung des Bluts. Klinische Wochenschr. **11**, 1375 72, 122, 151, 152, 180, 181, 190

—, 1934, Physico-chemical test for mitogenetic (GURWITSCH) rays. Nature **134**, 701 89

—, 1935, Physico-chemical test for mitogenetic (GURWITSCH) rays. Acta Brevia Neerland. **5**, 15 89

HENRICI, A. T., 1928, Morphologic Variation and the Rate of Growth of Bacteria. Springfield, C. C. Thomas. 185 pp. 120, 134

HERLANT, M., 1918, Periodische Änderungen der Permeabilität beim befruchteten Seeigelei. Compt. rend. soc. biol. **81**, 151 142

Page

Heyfetz see Braunstein.

Hill, J. H., and E. C. White, 1933, Action of normal skin on bacteria. Archives of Surgery **26**, 901 185

Himmelberger see Giltner.

Hollaender see Duggar.

Holzmann see Blacher.

Hughes, A. L., and L. A. du Bridge, 1932, Photoelectric Phenomena. New York, McGraw-Hill, 531 pp. 20, 26

Irichimowitsch see Blacher.

Jaeger, C., 1930, Der Einfluß der Blutstrahlung auf die Gewebskulturen. Z. f. Zellforschung **12**, 354 83

Jahn see Richards.

Jones, L. A., and V. C. Hall, 1926, On relation between time and intensity in photographic exposure (IV). J. Opt. Soc. Am. **13**, 460 . 24

Julius, H. W., 1935, Do mitogenetic rays have any influence on tissue cultures? Acta Brevia Neerland. **5**, 51 83

Kalendaroff, G. S., 1932, Die Spektralanalyse der Strahlung des markhaltigen Nerven im Ruhezustande und bei künstlicher Erregung. Pflügers Archiv **231**, 239 . . . . 73, 122, 155

Kannegiesser, N., 1931, Die mitogenetische Spektralanalyse I. Biochem. Z. **236**, 415 35

— see also Billig and Chariton.

Karpass, A. M., and M. N. Lanschina, 1929, Mitogenetische Strahlung bei Eiweißverdauung. Biochem. Z. **215**, 337 52

Kawaguchi, S., 1930, Über den Einfluß der Lichtstrahlen auf den Gesamt-Cholesteringehalt der Haut. Biochem. Z. **221**, 232 . . 183

Kendall see Kostoff

Kisliak-Statkewitsch, M., 1927, Die mitogenetische Strahlung des Kartoffeleptoms. Roux' Archiv **109**, 283 . . . . 141, 145

—, 1929, Die mitogenetische Strahlung des Carcinoms. Z. f. Krebsforschung **29**, 214 178

— see also Sorin.

Klee-Rawidowicz see Lasnitzky.

Klenitzky, J., 1932, Die mitogenetische Strahlung der weißen Blutelemente. Biochem. Z. **252**, 126 151

—, 1934, Le rétablissement de la radiation mitogénétique du sang après l'extirpation d'une tumeur cancéreuse. Arch. Sciences Biol. (Russ.) **35**, 218 181

— and E. Prokofiewa, 1934, L'analyse spectrale de la radiation mitogénétique de la dèsaggregation des polysaccharides. Arch. Sciences Biol. (Russ.) **35**, 211 39

Korösi, K. de, 1934, L'action à distance de la levure sur les sucres. I. Internat. Congress of Electro-Radio-Biology, Venice 45

Page

Kostoff, D., and J. Kendall, 1932, Origin of a tetraploid shoot from the region of a tumor on tomato. Science **76**, 144 169

Kreuchen, K. H., 1935, Messung geringer Lichtintensitäten mit Hilfe von Zählrohren. Z. f. Physik **94**, 549 92

— und J. B. Bateman, 1934, Physikalische und biologische Untersuchungen über mitogenetische Strahlung. Protoplasma **22**, 243 . . . . 92, 95, 190

— see also Hauser.

Kurepina see G. Frank.

Lanschina see Karpass.

Latmanisowa, L. W., 1932, Die mitogenetische Sekundärstrahlung des Nerven. Pflügers Archiv **231**, 265 . . . . . 110, 112, 113, 159

—, 1933, Parabiose der Nerven als Folge mitogenetischer Bestrahlung. Naturwissenschaften **21**, 330 159

—, 1934, Sur la parabiose mitogénétique du nerf. Ann. de Physiol. et de Physicochem. Biol. **10**, 141 159

Lasnitzki, A., and E. Klee-Rawidowicz, 1931, Zur Frage der mitogenetischen Induktion von Warmblüterzellen. Z. f. Krebsforschung **34**, 518 83

Lepeschkin, W. W., 1932a, Influence of visible and ultraviolet rays upon the stability of living matter. Am. J. Bot. **19**, 547 . . . 99

—, 1932b, Nekrobiotische Strahlen. Ber. d. deutsch. bot. Ges. **50**, 367 99

—, 1933, Nekrobiotische Strahlen I. Protoplasma **20**, 232 99

Loos, W., 1930, Untersuchungen über mitogenetische Strahlen. Jahrb. f. wissensch. Botanik **72**, 611 57

Liosner see Blacher.

Lorenz, E., 1933, Investigation of mitogenetic radiation by means of a photoelectric counter tube. U.S. Health Reports **48**, 1311 . 91, 188

—, 1934, Search for mitogenetic radiation by means of the photoelectric method. Journ. General Physiol. **17**, 843 . . . . . . 41, 91

Lucas, G. H. W., 1924, The fractionation of bios. Journ. physical Chem. **28**, 1180 73

Lubin see Macht.

Macht, D. I., and D. S. Lublin, 1923/24, A phyto-pharmacological study of menstrual toxin. Journ. of Pharm. and Exp. Ther. **22**, 413 . . . . 95, 184, 185

Magrou, J., and M. Magrou, 1927a, Recherches sur les radiations mitogénétiques. Bull. d'Histologie appliqué **4**, 253 . . . . 57, 178

—, —, 1927b, Radiations emises par le *Bacterium tumefaciens*. Rev. path. vég. et ent. agr. **14**, 244 . . . . 57, 178

—, —, 1927c, Rayons mitogénétiques et génése des tumeurs. Compt. rend. **184**, 905 . . . . 171, 178

—, —, 1928, Action à distance du *Bacterium tumefaciens* sur le développement de l'oeuf d'oursin. Compt. rend. **186**, 802 86

Page

MARGOU, J., and M. MARGOU, 1931, Action à distance de facteurs biologiques et chimiques sur le développement de l'oeuf d'oursin (*Paracentrotus lividus* L. K.). Annales des Sciences naturelles: Zoologie **14**, 149 . . . . 86, 165

—, **1932**, Action à distance et embryogénèse. Radiobiologia **1** (1), 32 86

— et P. REISS, 1931, Action à distance sur le développement de l'oeuf d'oursin. Essai d'interprétation. Compt. rend. Acad. Paris **193**, 609 86

MALCZYNSKI, S., 1930, Über den Einfluß der einmaligen Bestrahlung mittels Quarzlampe auf den Cholesteringehalt im Blute der nichtkrebsigen und krebskranken Personen. Klinische Wochenschrift **9**, 936 183

MAXIA, C., 1929, Intensificazione delle segmentazione din ova di *Paracentrotus lividus* sotto l'influenza di radiazione mitogenetiche. R. Com. Tallasografico It. Mem. **155** 142

— see also BRUNETTI.

MEES, C. E. K., 1931, Photographic plates for use in spectroscopy and astronomy. Journ. Opt. Soc. Am. **21**, 753 24

MOISSEJEWA, M., 1931, Zur Theorie der mitogenetischen Strahlung.
I. Biochem. Z. **241**, 1.
II. Biochem. Z. **243**, 67.
III. Biochem. Z. **251**, 133 . . . . 62, 188

NAKAIDZUMI, M., und H. SCHREIBER, 1931, Untersuchungen über das mitogenetische Strahlungsproblem II. Biochem. Z. **237**, 358 70, 190

NAKAJIMA see WRIGHT.

NAUMANN, H., 1919, Die Lebenstätigkeit von Sproßpilzen in mineralischen Nährlösungen. Z. f. techn. Biol. **7**, 1 138

NEBLETTE, C. B., 1930, Photography. 2nd ed. New York, Van Nostrand. 198 pp.. 116

NELSON, C. and S. C. BROOKS, 1933, Effect of infra-red light on subsequent fertilisation of the eggs of certain marine invertebrates. Proc. Soc. Exp. Biol. and Med. **30**, 1007 101

NORTH, C. E., 1909, Reports of 300 cases treated with lactic acid bacteria. Medical Record, March 27 177

OKUNEFF, N., 1928, Über einige physico-chemische Erscheinungen während der Regeneration. Biochem. Z. **195**, 421 174

OUELLET see GREY.

PAUL, M., 1933, Zwiebelwurzeln als Detektoren bei Untersuchungen über mitogenetische Strahlung. Roux' Archiv **128**, 108 57, 62, 189

PENFOLD, W. J., 1914, On the nature of bacterial lag. J. Hyg. **14**, 215 134

PFEIFFER, G., 1928—31, Die Cholesterine im Strukturverbande des Protoplasmas. Biochem. Z. **201**, 424; **220**, 53 and 210; **222**, 214; **230**, 439; **231**, 239; **232**, 255; **235**, 97; **236**, 457 186

Page

Polano, O., and K. Dietl, 1924, Die Einwirkung der Hautabsonderung bei der Menstruierenden auf die Hefegärung. Münchener Med. Wochenschrift **71**, 1385 95

Ponomarewa, J., 1931, Das detaillierte glykolytische Spektrum. Biochem. Z. **239**, 424 37

Potozky, A., 1930, Über Beeinflussung des mitogenetischen Effekts durch sichtbares Licht. Biol. Zentr. **50**, 712 108

—, 1932, Die mitogenetischen Spektra der Oxydationsreaktionen. Biochem. Z. **249**, 282 36

—, S. Salkind and I. Zoglina, 1930, Die mitogenetische Strahlung des Bluts und der Gewebe von Wirbellosen. Biochem. Z. **217**, 178 146

— and I. Zoglina, 1928, Über mitogenetische Sekundärstrahlung aus abgeschnittenen Zwiebelwurzeln. Roux' Archiv **114**, 1 . 109

— and I. Zoglina, 1929, Mitogenetische Strahlung des Bluts. Biochem. Z. **211**, 352 149

— see also Braunstein and Salkind.

Prokofiewa see Klenitzky.

Protti, G., 1930, Impressioni fotografiche di radiazioni ematiche ottenuti attraverso il quarzo. Comm all Sed Scient. Osped. Civile di Venezia 90

—, 1931, L'emoinnesto intramuscolare. Milano, Ulrico Hoepli. 140 pp. 151

—, 1934a, Il fenomeno della emoradiazione applicato alla clinica. Radiobiologia **1** (4), 49 153

—, 1934b, Das Verhalten einiger Tumoren gegenüber dem Saccharomyces cerevisiae. Arch. Sciences Biol. (Russ.) **35**, 255 see also: Effetti citolitici da radiazioni biologiche (citofotolisi). La Ricerca Scientifica **2**, No 99 182

Rahn, Otto, 1906, Über den Einfluß der Stoffwechselprodukte auf das Wachstum der Bakterien. Centr. f. Bakt. II. Abt. **16**, 418 134

—, 1929, The fermentometer. Journ. Bact. **18**, 199 125

—, 1932, Physiology of Bacteria. Philadelphia, Blakiston. 427 pp. 131

— and M. N. Barnes, 1933, An experimental comparison of different criteria of death in yeast. Journ. Gen. Physiol. **16**, 579 . . . 125

— see also Barnes and Ferguson and Tuthill.

Rajewsky, B., 1931, in Fr. Dessauer „Zehn Jahre Forschung auf dem physikalisch-medizinischen Grenzgebiet". Leipzig 1931 . 91

—, 1932, Zum Problem der mitogenetischen Strahlung. Z. f. Krebsforschung **35**, 387 91

—, 1934, Über einen empfindlichen Lichtzähler. Physik. Z. **32**, 121 . 92

Ras see Wolff.

Reiter, I., and D. Gabor, 1928, Zellteilung und Strahlung. Sonderheft der Wissenschaftl. Veröffentl. aus d. Siemens-Konzern. Berlin, J. Springer . . . 57, 59, 81, 90, 120, 129, 148, 155, 169

Reiss see Margou.

Richards, O. W., and T. L. Jahn, 1933, A photoelectric nephelometer. Journ. Bacteriol. **26**, 385 74

— and G. W. Taylor, 1932, „Mitogenetic Rays“ — a critique of the yeast detector method. The Biol. Bulletin **63**, 113 69

Rivers, T. M., and F. L. Gates, 1928, Ultraviolet light and vaccine virus. Journ. Exper. Med. **47**, 45 48

Robertson, T. B., 1924, The nature of the factors which determine the duration of the period of lag in cultures of infusoria. Austral. Journ. Exp. Biol. and Med. Sci. **1**, 105 137

— Influence of washing etc. Ibid. **1**, 151 137

Rodionow see G. Frank.

Roffo, A. H., 1933, Heliotropism of cholesterol in relation to skin cancer. Am. Journ. of Cancer **17**, 42 182

—, 1934a, Héliotropisme de la cholesterine. I. Internat. Congress of Electro-Radio-Biology, Venice 183

—, 1934b, Action cancérigène des irradiations solaires. I. Internat. Congress of Electro-Radio-Biology, Venice 183

Rossmann, B., 1928, Untersuchungen über die Theorie der mitogenetischen Strahlen. Roux' Archiv **113**, 346 . . . . 57, 58

Rubner, Max, 1912, Die Ernährungsphysiologie der Hefezelle bei der alkoholischen Gärung. Archiv f. Physiol., Suppl. 1912, p. 1 . 41

Runnström, J., 1928, Struktur und Atmung, bei der Entwicklungserregung des Seeigeleies. Acta zoolog. **9**, 445 85

Ruyssen, R., 1933, L'émission de rayons de Gurwitsch par les réactions chimiques entre gaz. Acad. Roy. Belgique, Sciences, 5e série, **19**, 535 40

Saenger, H., 1921, Gibt es ein Menstruationsgift? Zentr. f. Gynäkol. **45**, 819 95

Salkind, S., 1933, Beiträge zur Analyse der mitogenetischen Effekte, I. Roux' Archiv **128**, 378 . . . . 70, 114, 116

—, 1931, Die mitogenetische Strahlung der Larve von *Saccocirrus*. Roux' Archiv **124**, 467 144

—, A. Potozky and I. Zoglina, 1930, Die mitogenetische Beeinflussung der Eier von *Protodrilus* und *Saccocirrus*. Roux' Archiv **121**, 630 . . . . 81, 143

— und J. Ponomarewa, 1934, Der unmittelbare Einfluß der mitogenetischen Strahlen auf den Verlauf der Zellteilung. Radiobiologia **1**, (4), 3 191

Page

Salkind see also G. Frank, L. Gurwitsch and Potozky.

Samarajew, W. N., 1932, Die mitogenetische Ausstrahlung bei der Regeneration des Regenwurms. Roux' Archiv **126**, 633 . . . 175

— see also Blacher.

Schick, B., 1920, Das Menstruationsgift. Wiener klin. Wochenschr. **33**, 395 . . . 95

Schouten, S. L., 1933, Gurwitsch-Strahlen und Pilzsporenkeimung. Acta Brevia Neerland. **3**, 68 . . . 80

Schreiber, H., 1933, Zur Theorie der „mitogenetischen Strahlung". Protoplasma **19**, 1 . . . 79

— und W. Friedrich, 1930, Über Nachweis und Intensität der mitogenetischen Strahlung. Biochem. Z. **227**, 386 . . . 92

— see also Nakaidzumi.

Schwarz, 1928, Zur Theorie der mitogenetischen Strahlung. Biol. Zentralbl. **48**, 302 . . . 57, 58

Schwemmle, J., 1929, Mitogenetische Strahlen. Biol. Zentralbl. **49**, 421 . . . 57, 190

Scott, L. C., 1931, The determination of potassium in cardiac muscle and the presumable influence of the beta-radiations on the rythm. Journ. of Clinical Med. **10**, 745 . . . 102

Severin see Braunstein.

Sewertzowa, L. B., 1929, Über den Einfluß der mitogenetischen Strahlen auf die Vermehrung der Bakterien. Biol. Zentralbl. **49**, 212 . . . 75, 78

Seyderhelm, R., 1932, Über einen durch ultraviolette Bestrahlung aktivierbaren, antianämisch wirkenden Stoff im Blut. Klinische Wochenschr. **11**, 628 . . . 151

Siebert, W. W., 1928a, Über die mitogenetische Strahlung des Arbeitsmuskels und einiger anderer Gewebe. Biochem. Z. **202**, 115 . . . 64, 147

—, 1928b, Über die Ursachen der mitogenetischen Strahlung. Biochem. Z. **202**, 133 . . . 64, 71

—, 1929, Aktionsstrahlung des Muskels und Wachstumswirkung des elektrodynamischen Feldes. Biochem. Z. **215**, 152 . . . 71

—, 1930, Die mitogenetische Strahlung des Bluts und des Harns gesunder und kranker Menschen. Biochem. Z. **226**, 253 65, 71, 153

—, 1934, Die „mitogenetische" Strahlung des Bluts in Handb. d. allg. Hämatologie Bd. II, 2, 1339. Berlin, Urban und Schwarzenberg . . . 148

— und H. Seffert, 1933, Physikalischer Nachweis der Gurwitsch-Strahlung mit Hilfe eines Differenzverfahrens. Naturwissenschaften **21**, 193 . . . 91

—, —, 1934, Zur Frage des physikalischen Nachweises der Gurwitsch-Strahlung. Archiv Sciences Biol. (Russ.) **35**, 177 . . . 91

Page

Solowjeff see Billig.

Sorin, A. N., and M. Kisliak-Statkewitsch, 1928, Über mitogenetische Induktion in den frühen Entwicklungsstadien des Hühnerembryo. Roux' Archiv **113**, 724. 145

Stempell, W., 1929, Nachweis der von frischem Zwiebelsohlenbrei ausgesandten Strahlen durch Störung der Liesegangschen Ringbildung. Biol. Zentralbl. **49**, 607 88

—, 1931, Das Wasserstoffsuperoxyd als Detektor für Organismenstrahlung und Organismengasung. Protoplasma **12**, 538 . 89, 101

—, 1932, Die unsichtbare Strahlung der Lebewesen. Jena, Gustav Fischer, 88 pp. 59, 89, 93, 164

Strauss, W., 1929, Objektive Nephelometrie mittels des Mollschen Trübungsmessers, demonstriert am Beispiel der Bakterienzählung. Centr. f. Bakt., I. Abt. Orig. **115**, 228 74

Suchow, K., and M. Suchowa, 1934, Über Coagulationsstrahlung. Archiv Sciences Biol. (Russ.) **35**, 307 100

Sussmanowitsch, H., 1928, Erschöpfung durch mitogenetische Induktion. Roux' Archiv **113**, 753 115

Taylor, G. W., and E. N. Harvey, 1931, The theory of mitogenetic radiation. Biol. Bulletin **61**, 280 . . . . 57, 58

— see also Richards.

Taylor, H. S., 1931, A Treatise on Physical Chemistry. New York, D. van Nostrand Co. 46, 90

Tuthill, J. B., and Otto Rahn, 1933, Zum Nachweis mitogenetischer Strahlung durch Hefesprossung. Archiv f. Mikrobiol. **4**, 565 66, 120, 121, 190

Unna, P. G., and L. Golodetz, 1909, Die Hautfette. Biochem. Z. **20**, 469 186

Wagner, N., 1927, Über den von A. Gurwitsch entdeckten spezifischen Erreger der Zellteilung (mitogenetische Strahlen). Biol. Zentralbl. **47**, 670 . . . . 57, 58

Warburg, Otto, 1909, Über die Oxydationen im Ei. Z. Physiol. Chem. **60**, 443 142

—, 1919, Über die Geschwindigkeit der photochemischen Kohlensäurezersetzung in lebenden Zellen. Biochem. Z. **100**, 230 105

Wassilieff, L. L., 1934, De l'influence du travail cerebral sur la radiation mitogenetique du sang. Arch. Sciences Biol. (Russ.) **35**, 104 150

—, G. M. Frank und E. E. Goldenberg, 1931, Versuche über die mitogenetische Strahlung der Nerven. Biol. Zentralbl. **51**, 225 155

Werner, S., 1935, Die Entladungsformen im zylindrischen Zählrohr. Z. f. Physik **92**, 705 29

White see Hill.

Page

WILDIERS, E., 1901, Une nouvelle substance indispensible au développement de la lévure. La Cellule **18**, 313 138

WOLFF, L. K., and G. RAS, 1931, Einige Untersuchungen über die mitogenetischen Strahlen von GURWITSCH. Centr. Bakt. I Orig. **123**, 257 . . . . 75, 76, 190

—, —, 1932, Über mitogenetische Strahlen: II. Biochem. Z. **250**, 305 39, 121, 132

—, —, 1933 a, Über mitogenetische Strahlen: III. Biochem. Z. **259**, 210 76

—, —, 1933 b, Über mitogenetische Strahlen: IV. Über Sekundärstrahlung. Centr. f. Bakt. I Orig. **128**, 306. . . . . 43, 109

—, —, 1933 c, V.: Über die Methodik zum Nachweis der GURWITSCH-Strahlen. Centr. f. Bakt. I Orig. **128**, 314 . . . . 80, 93, 116, 146

—, —, 1934 a, VI.: Le rayonnement secondaire. K. Akad. Wetensch. Amsterdam **37**, 1 . . . . 45, 109, 113, 114, 123, 190

—, —, 1934 b, Effects of mitogenetic rays upon eggs of Drosophila. Nature **133**, 499. . . . . 81, 143

WORONZOWA see BLACHER.

WRIGHT, W. H., and H. NAKAJIMA, 1929, The growing of pure cultures from single cells of non-spore forming bacteria. Journ. Bact. **17**, 10 138

ZIRPOLO, G., Ricerche sulle radiazione mitogenetiche. Boll. Soc. di Naturalisti Napoli **42**, Atti 169 . . . . 81, 142

ZOGLINA see POTOZKY and SALKIND.

ZWAARDEMAKER, H., 1921, Über die Bedeutung der Radioaktivität für das tierische Leben. Ergebnisse d. Physiol. **19**, 326 102

—, 1926/27, Über das Erwachen des durch Kaliumentziehung zur Ruhe gekommenen Herzens durch die Bestrahlung des Radiums. Archiv f. d. ges. Physiol. **215**, 460 102

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